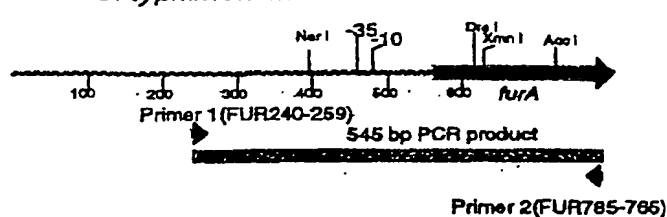


FIGURE 1A

S. typhimurium chromosomal DNA

Primer 1: FUR240-259
5'-TTT CGG CTG AAT TGC ACC TC-3'

Primer 2: FUR785-765
5'-CCT TCA AAA TTA TGG CGG GTC-3'

Primer 3: INV FUR380-400BGL
5'-GAA GAT CTC CAC CTG ATT ACA CCC GGAC-3'

Primer 4: INV FUR564-544NHE
5'-CGG CTA GCA ACA GGA CAG ATT CCG CATG-3'

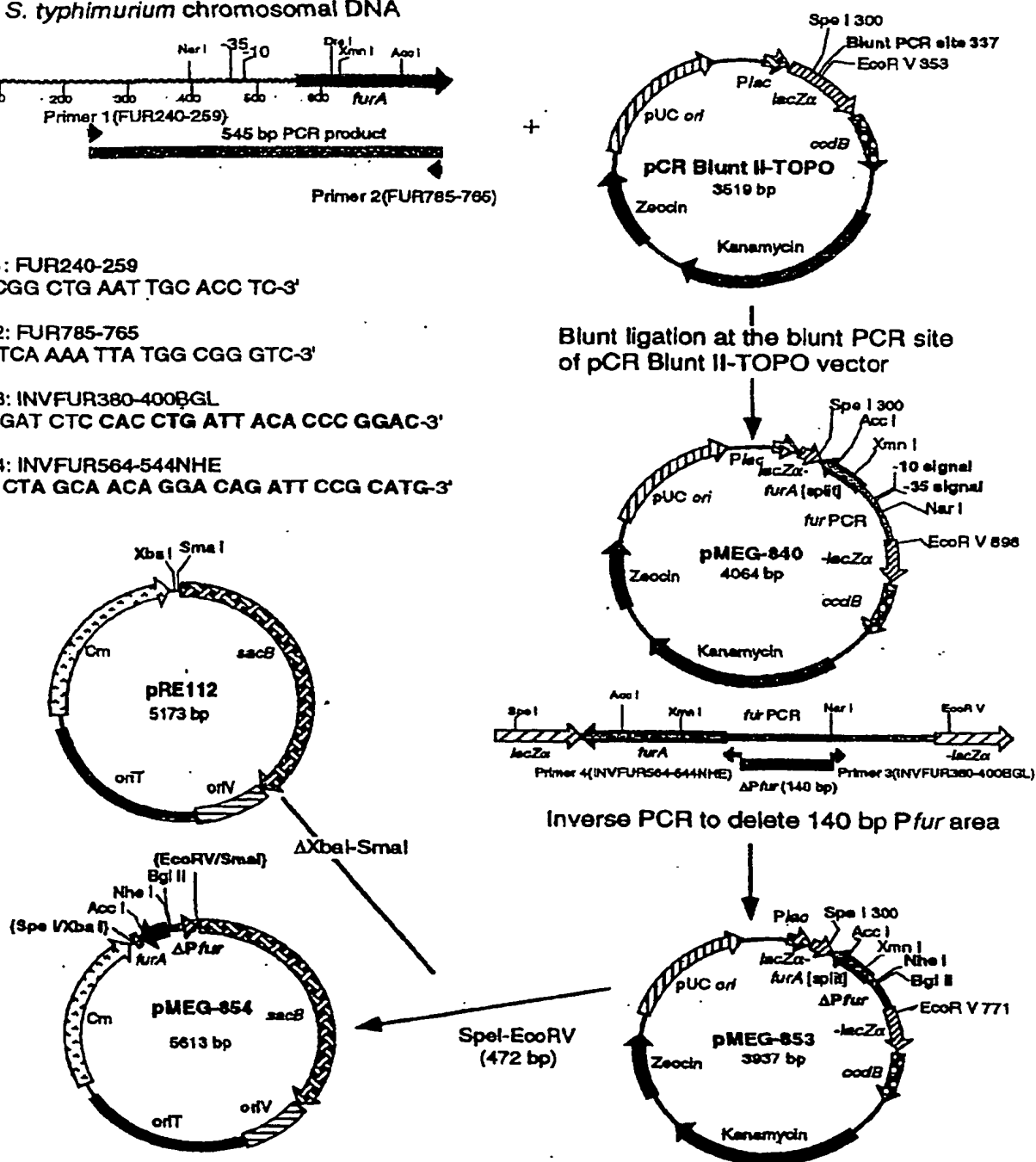


Figure 1-A. Construction of suicide vector for transfer of $\Delta P_{fur}223::T_{TaraC} P_{BAD} fur$ deletion-insertion mutation.

FIGURE 1-B

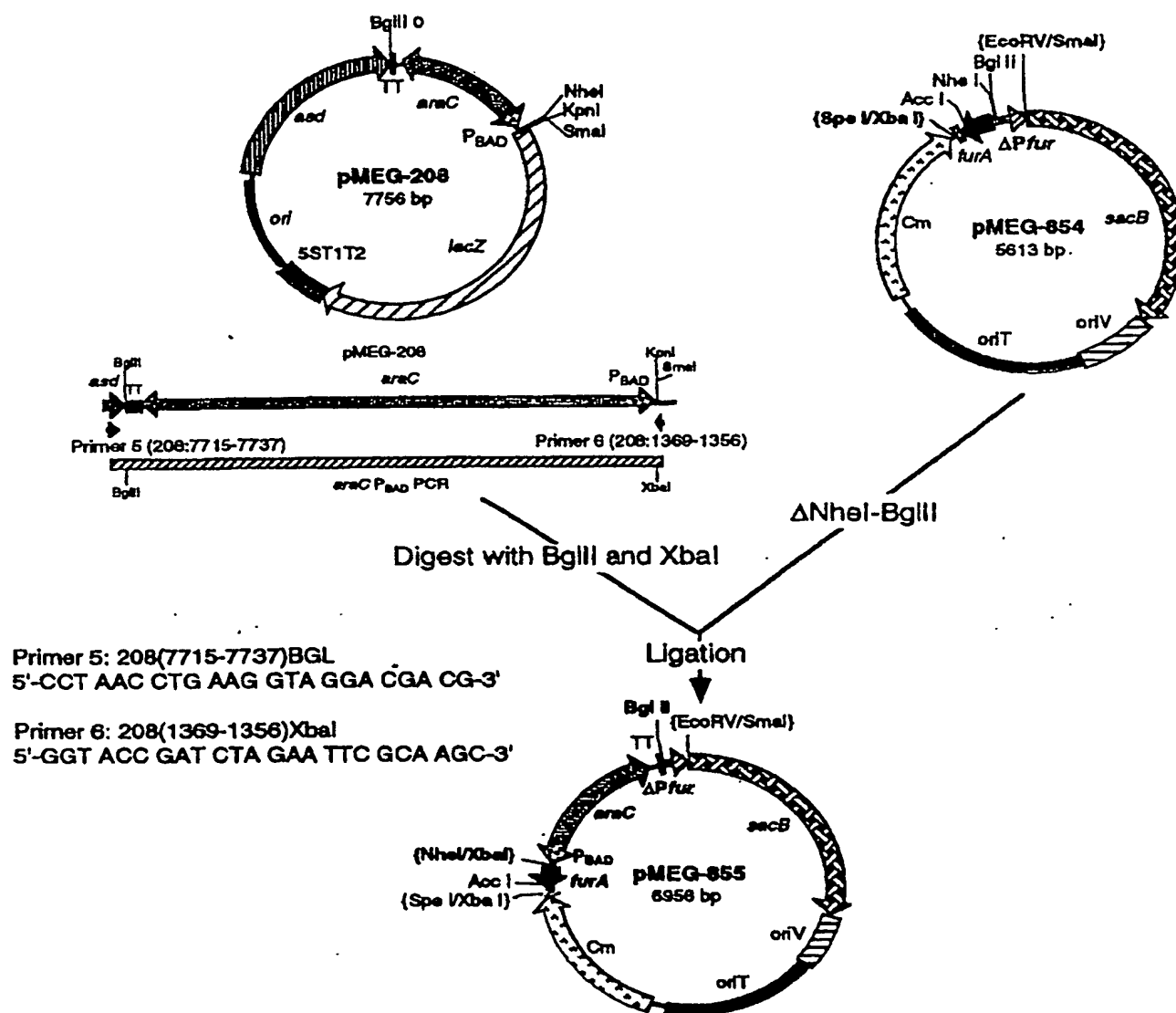
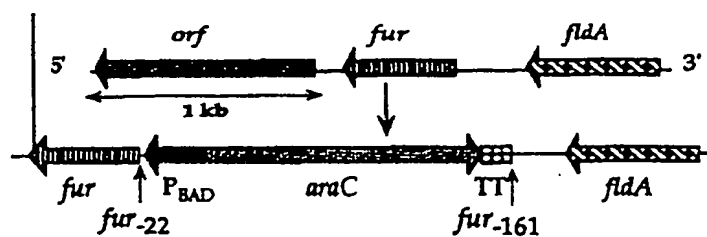


Figure 1-B. Construction of suicide vector for transfer of $\Delta P_{fur}223::TTaraC P_{BAD} fur$ deletion-insertion mutation.



140 bp *fur* promoter region deleted
1,354 bp TT *araC* P_{BAD} inserted

Figure 2. $\Delta P_{fur223}::TT_{araC} P_{BAD} fur$ deletion-insertion chromosomal construction.

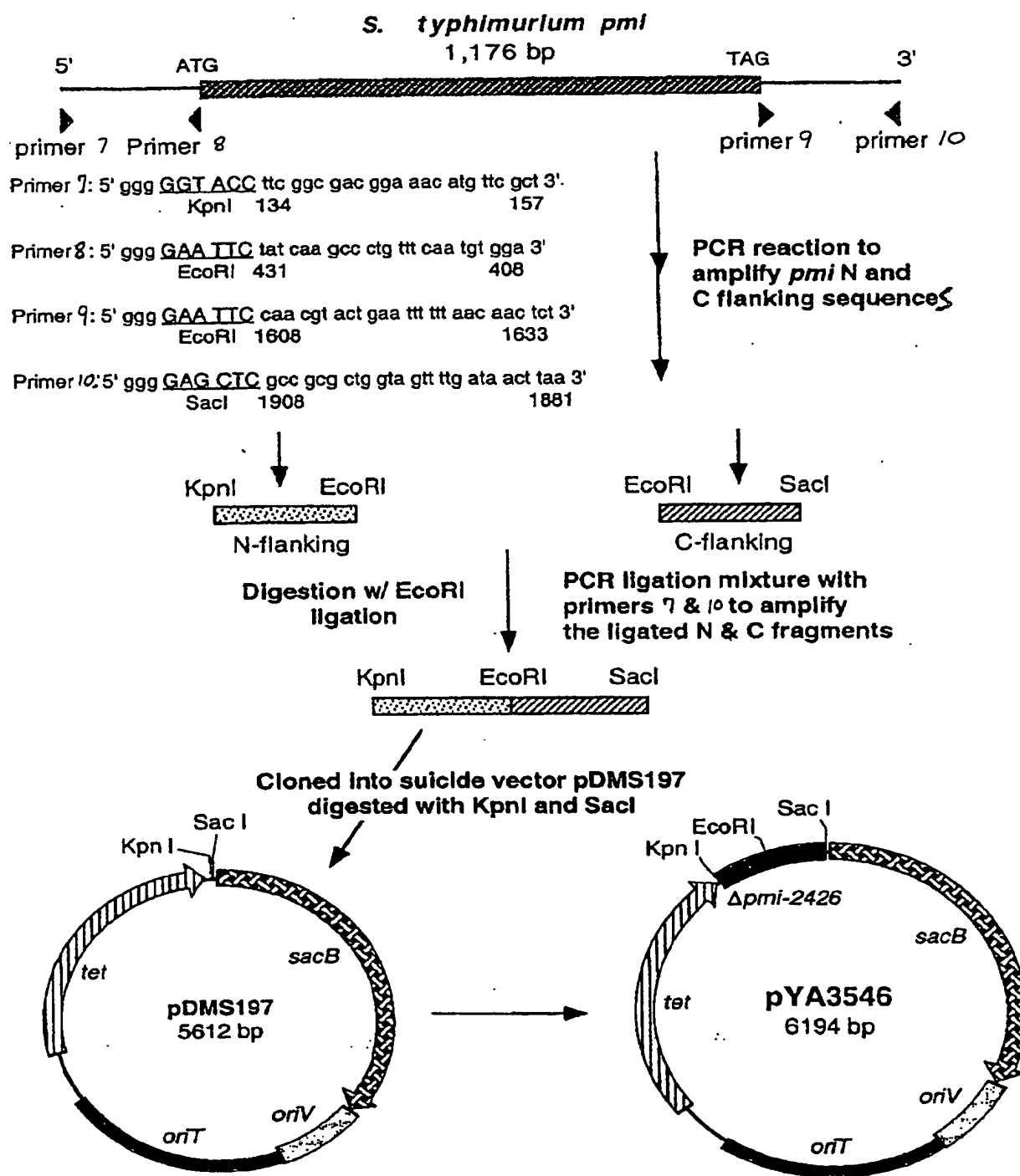
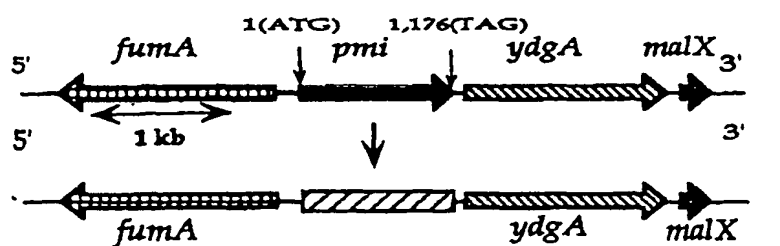


Figure 3. Construction of a suicide vector for *pmi* deletion,



1,176 bp *pmi* gene deleted (from ATG to TAG)

Figure 4. Chromosomal deletion for Δpmi -2426

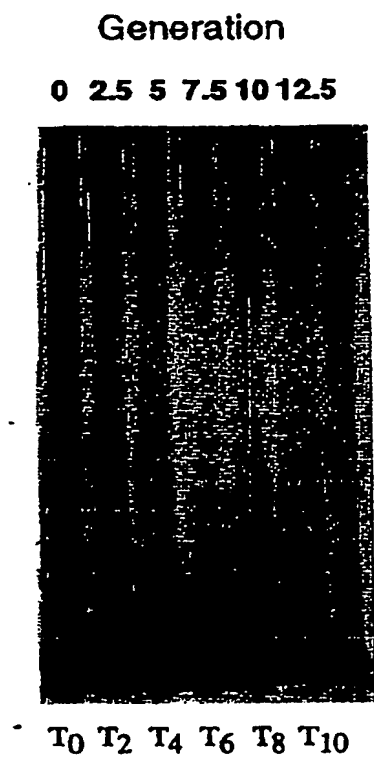
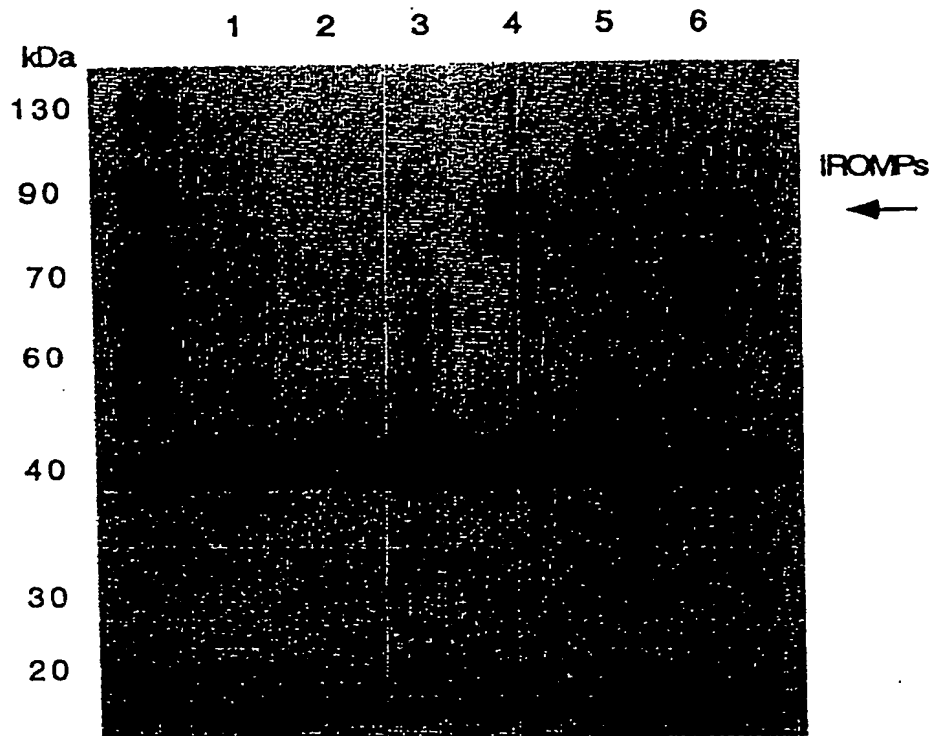


Figure 5. Reduction of LPS O-side chains in χ 8650 as a function of numbers of generations of growth or times (hours) of sampling.



Lanes:

- | | | |
|----------------|--|--------------------------------------|
| 1. χ 3761 | wild-type | Nutrient broth (NB) + 0.2% arabinose |
| 2. χ 3761 | wild-type | NB |
| 3. χ 8634 | $\Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur}$ | NB + 0.2% arabinose |
| 4. χ 8634 | $\Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur}$ | NB |
| 5. χ 8754 | $\Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur } \Delta pmi-2426$ | NB + 0.2% arabinose |
| 6. χ 8754 | $\Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur } \Delta pmi-2426$ | NB |

Figure 6. Outer membrane protein profile of $\Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur}$ mutants grown in Nutrient broth +/- arabinose.

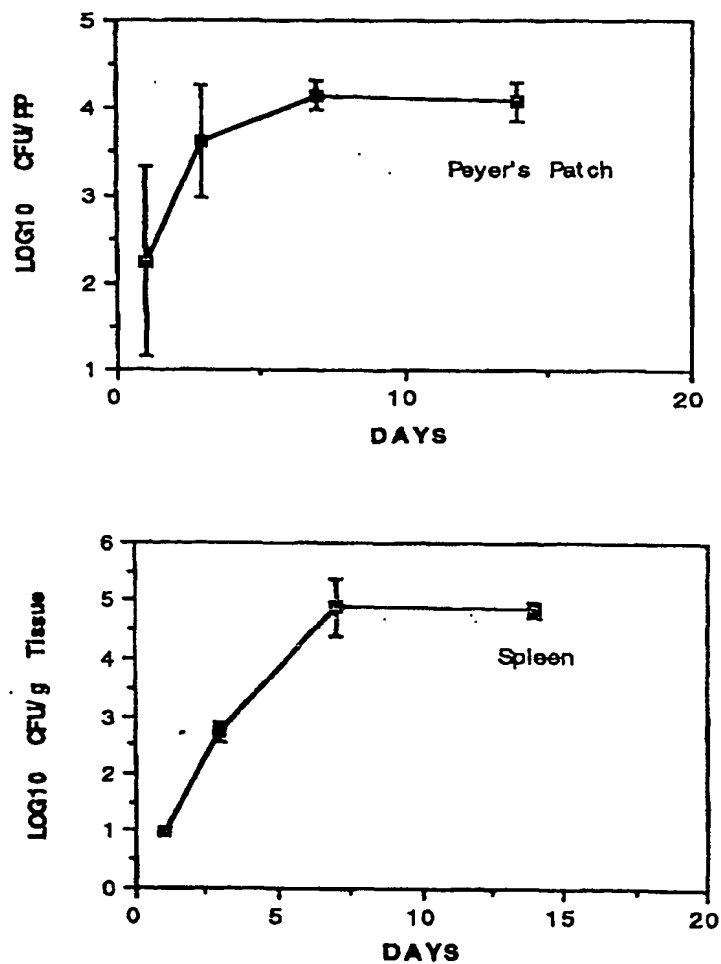


Figure 7. Colonization of 8-week-old female BALB/c mice with $\chi 8634 \Delta P_{fur} 223::T_{Tara} C P_{BAD} fur$ following oral inoculation.

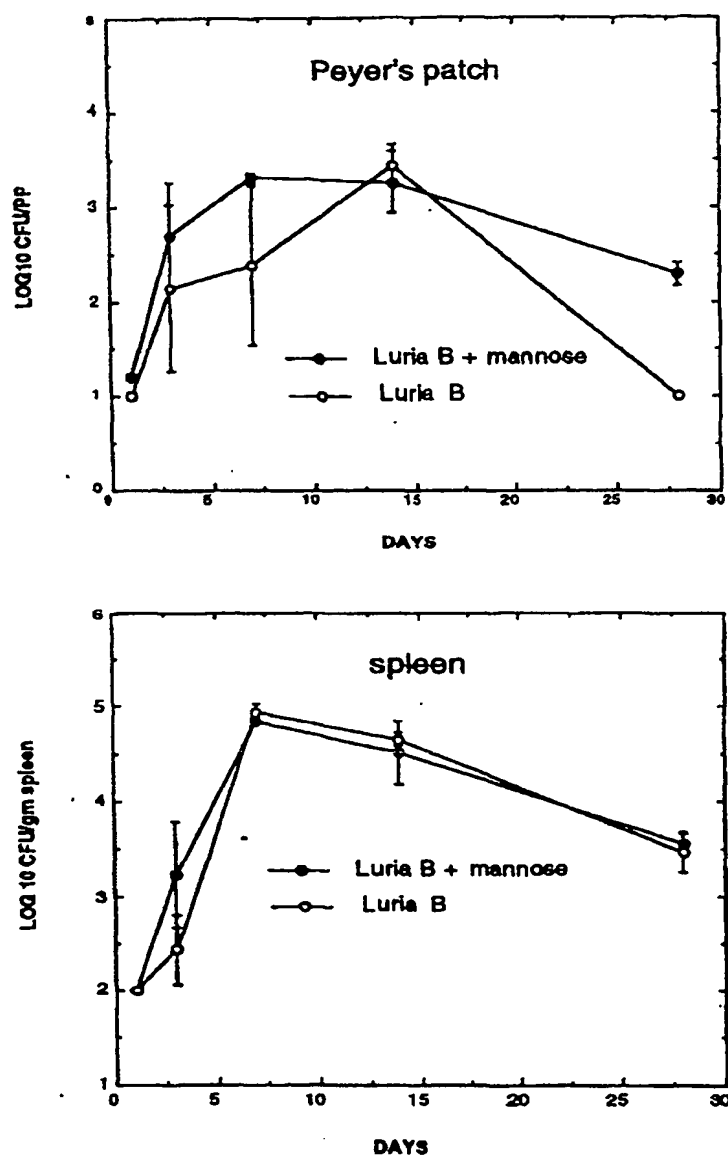


Figure 8. Colonization of 8-week-old female BALB/c mice with χ 8650 (Δpmi -2426) following oral inoculation.

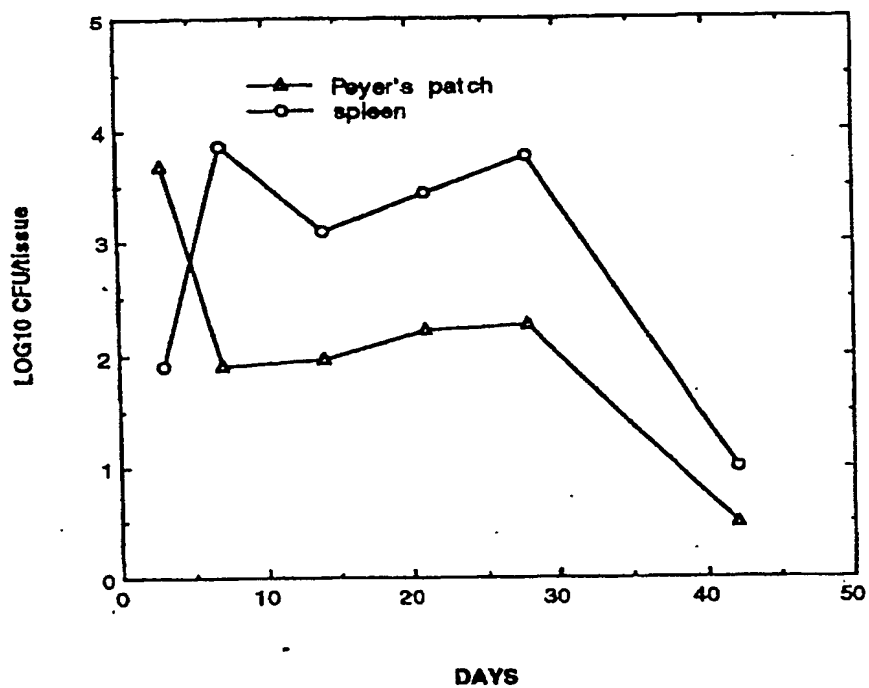


Figure 9. Colonization of 8-week-old female BALB/c mice with $\chi 8754$ ($\Delta pml-2426 \Delta P_{fur223}::araC P_{BAD} fur$) following oral inoculation.

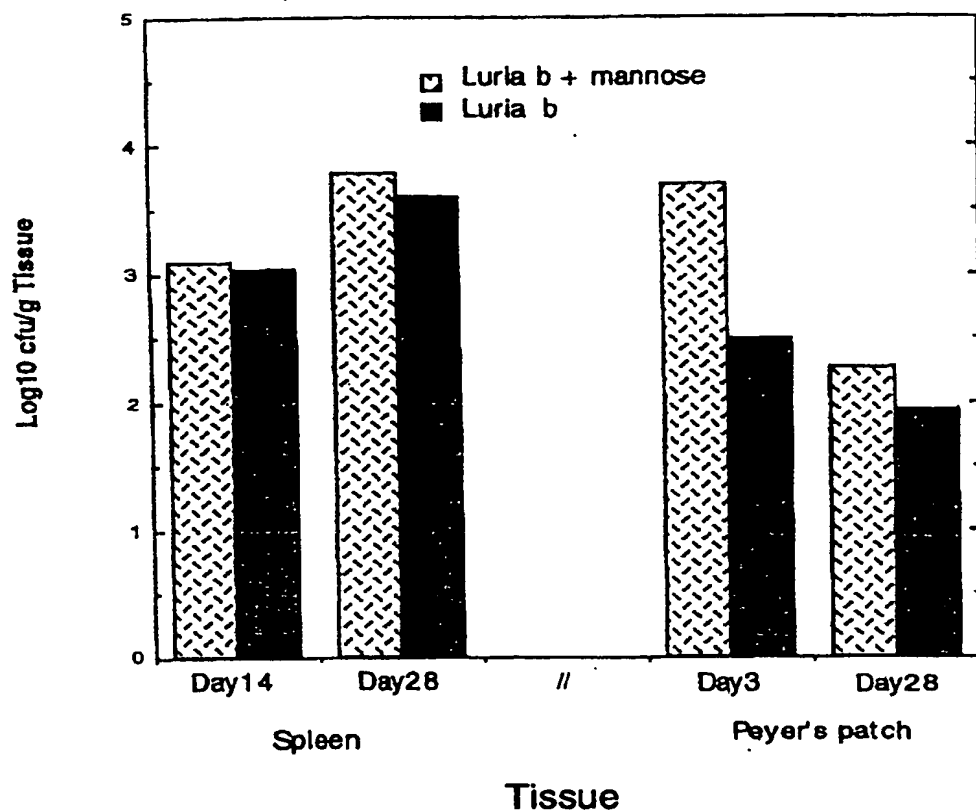
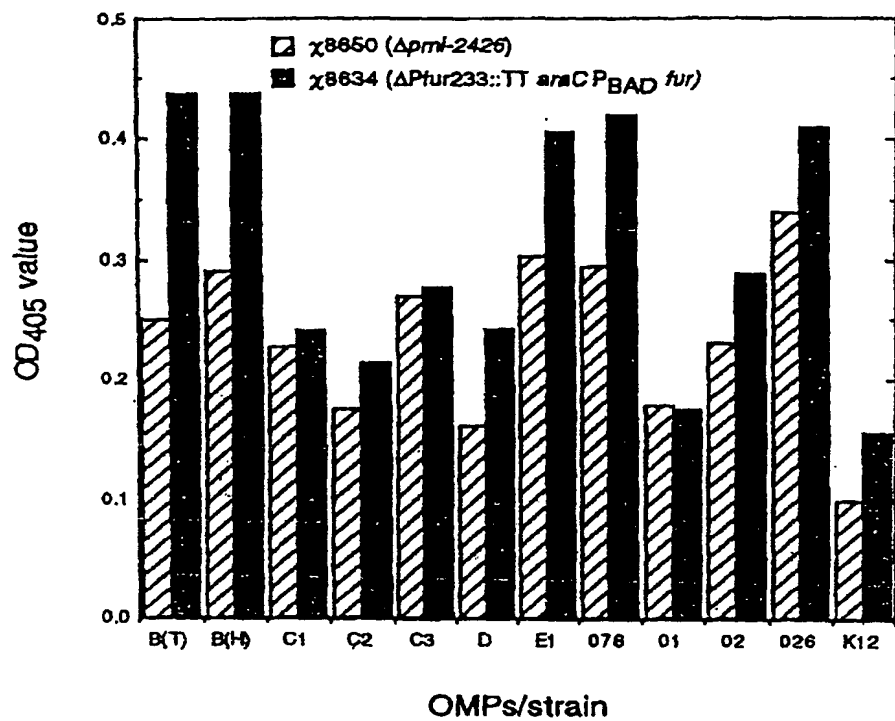


Figure 10. Colonization of 8-week-old female BALB/c mice with $\chi 8754$ ($\Delta pml-2426 \Delta P_{fur} 223::TTaraC P_{BAD} fur$) following oral inoculation.



B (T):	<i>S. typhimurium</i>	χ3761	E1:	<i>S. anatum</i>	χ4449
B (H):	<i>S. heidelberg</i>	χ3242	078:	APEC	χ7122
C1:	<i>S. infantis</i>	χ3212	01:	APEC	χ7237
C2:	<i>S. hadar</i>	χ3210	02:	APEC	χ7255
C3:	<i>S. albany</i>	χ3202	026:	EPEC	χ6206
D:	<i>S. enteritidis</i>	χ3700	K-12	<i>E. coli</i> K-12	χ289

Figure 11. IgG Ab responses to OMPs isolated from *Salmonella* and *E. coli* strains.

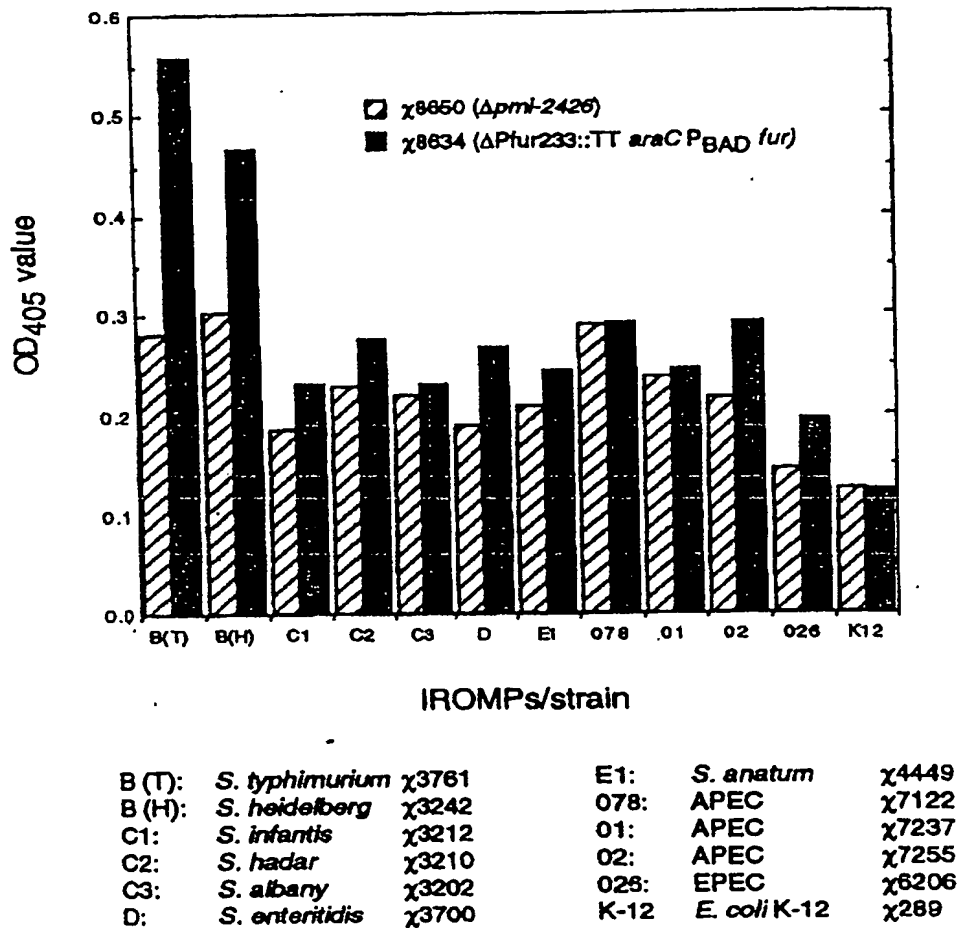


Figure 12. IgG Ab responses to IROMPs isolated from *Salmonella* and *E. coli* strains.

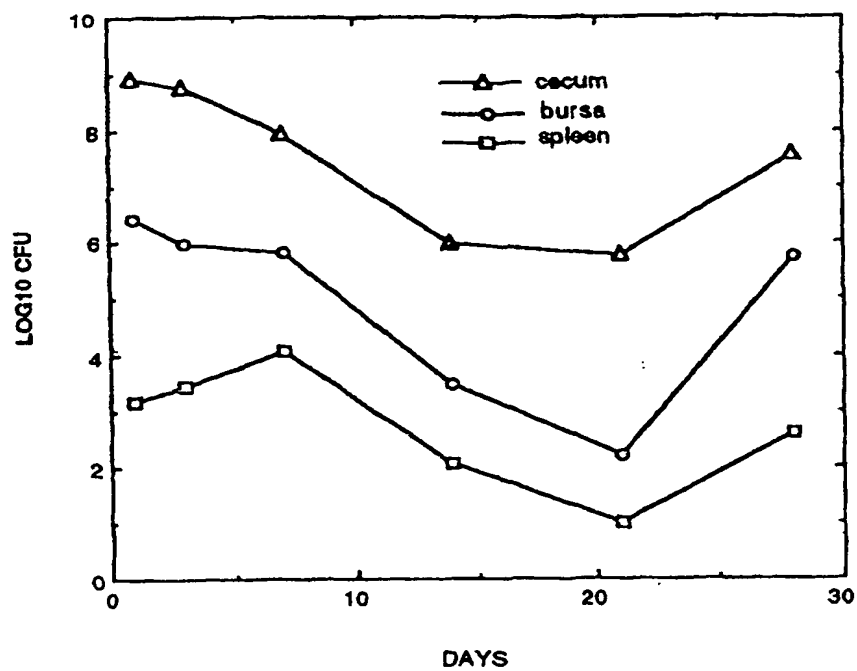


Figure 13. Colonization of day-of-hatch chicks with $\chi 8754$ ($\Delta pmi-2426 \Delta P_{fur223}::TT \text{ araC } P_{BAD} \text{ fur}$) following oral inoculation.

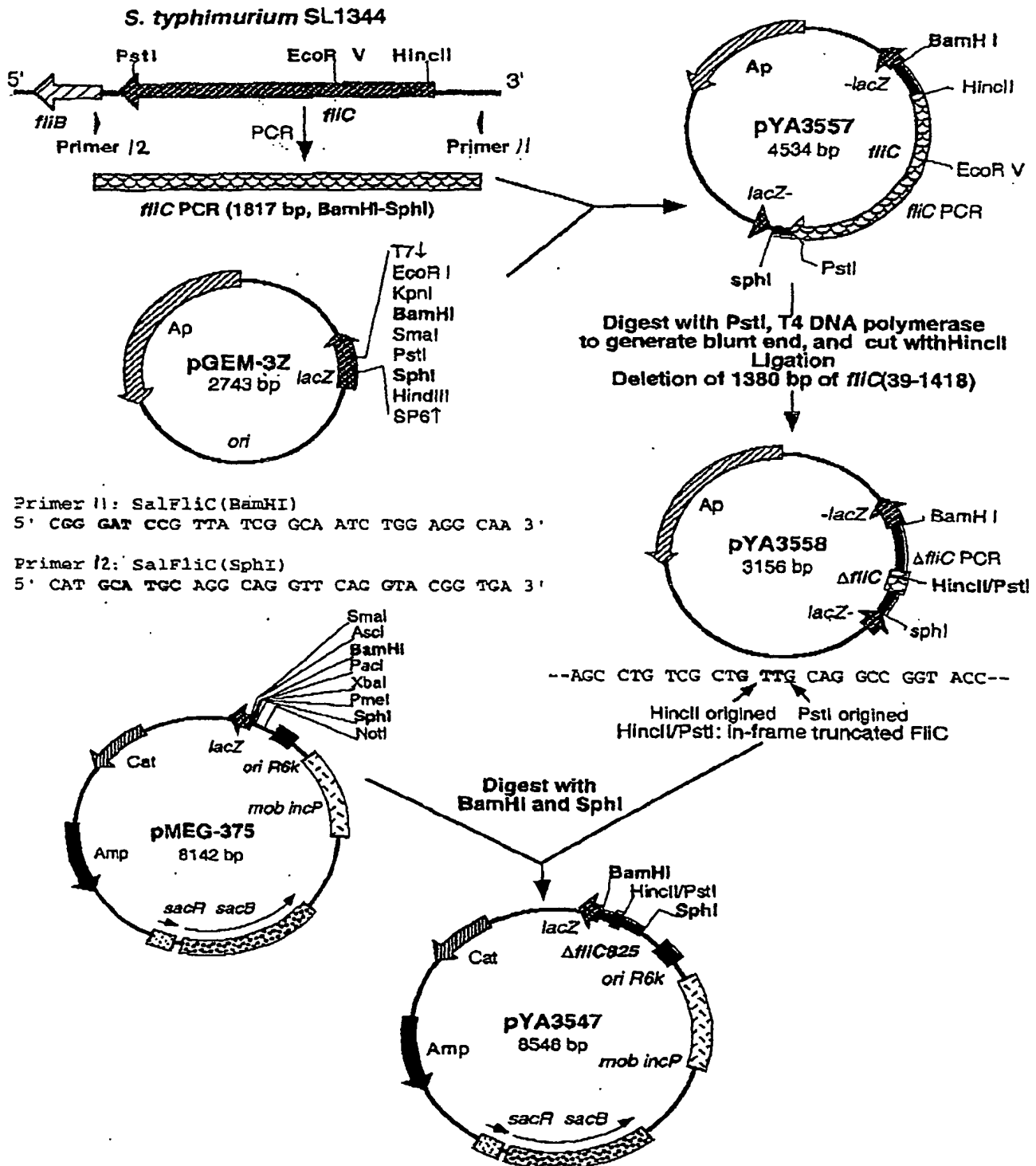
FIGURE 14. Construction of suicide vector for $\Delta fliC825$ 

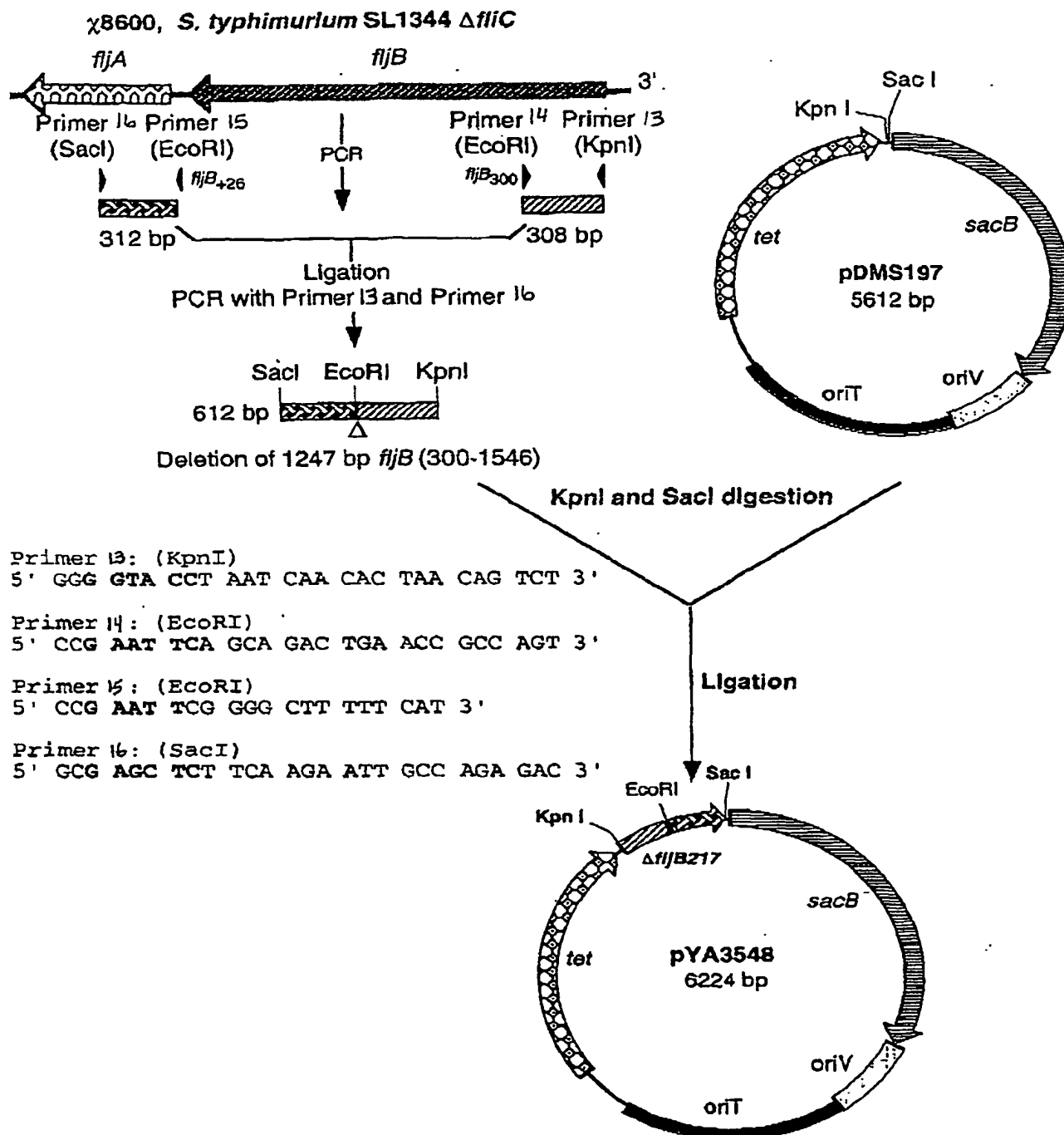
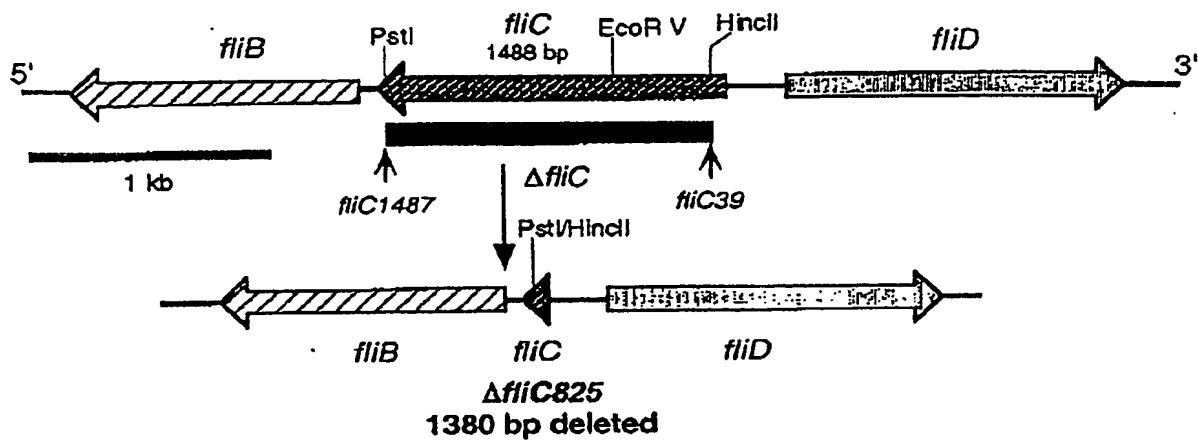
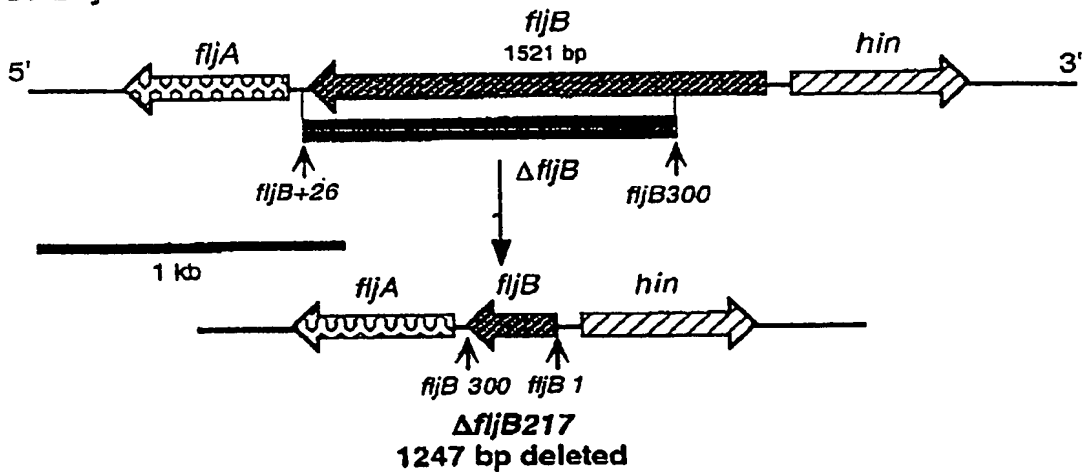
FIGURE 15. Construction of sulcid vector for $\Delta fljB217$ 

FIGURE 16. *Salmonella typhimurium* SL1344 chromosomal deletions:

A. $\Delta fliC825$



B. $\Delta fljB217$



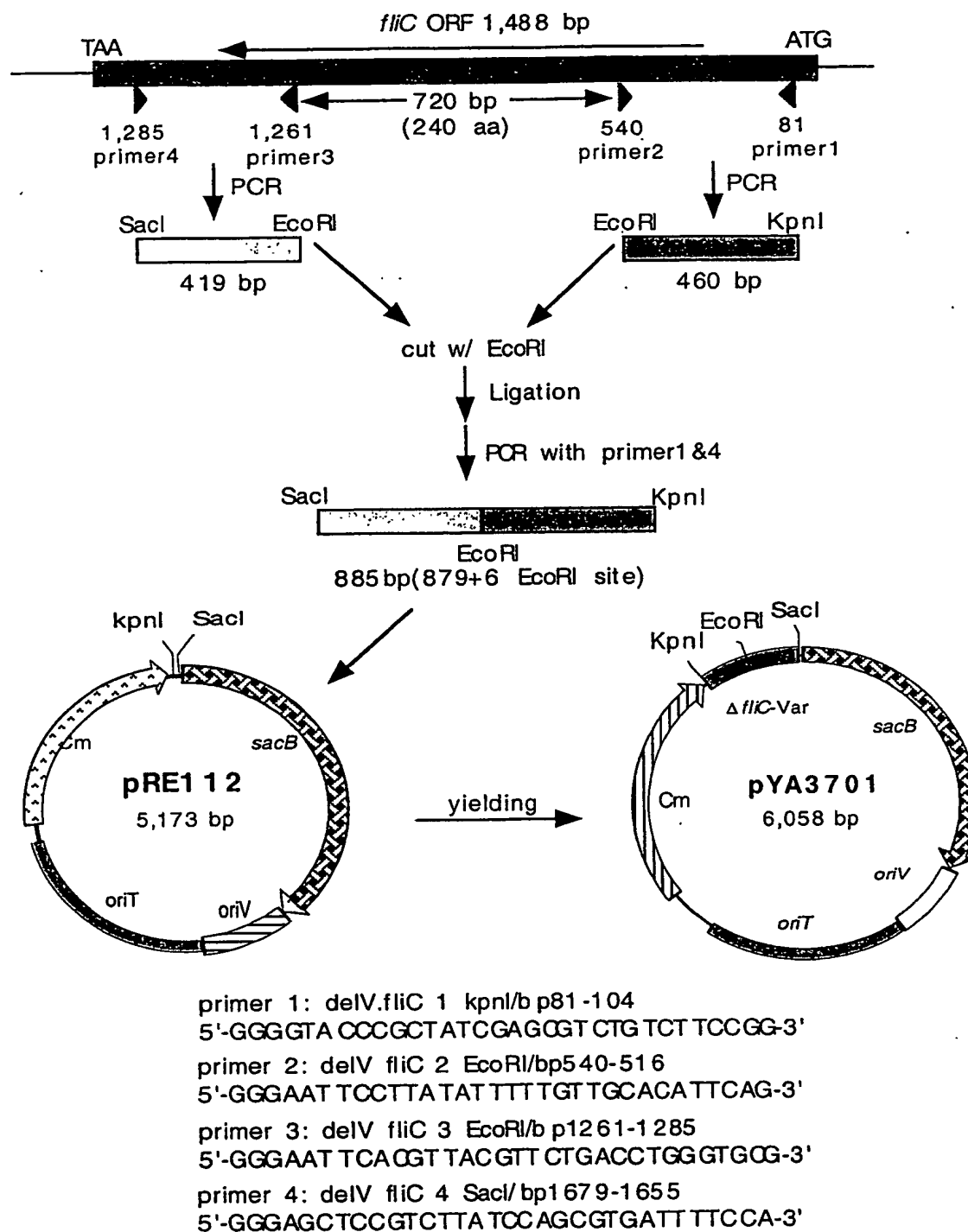


Figure 17. Construction of a suicide vector for transfer of $\Delta fliC$ -Var mutation

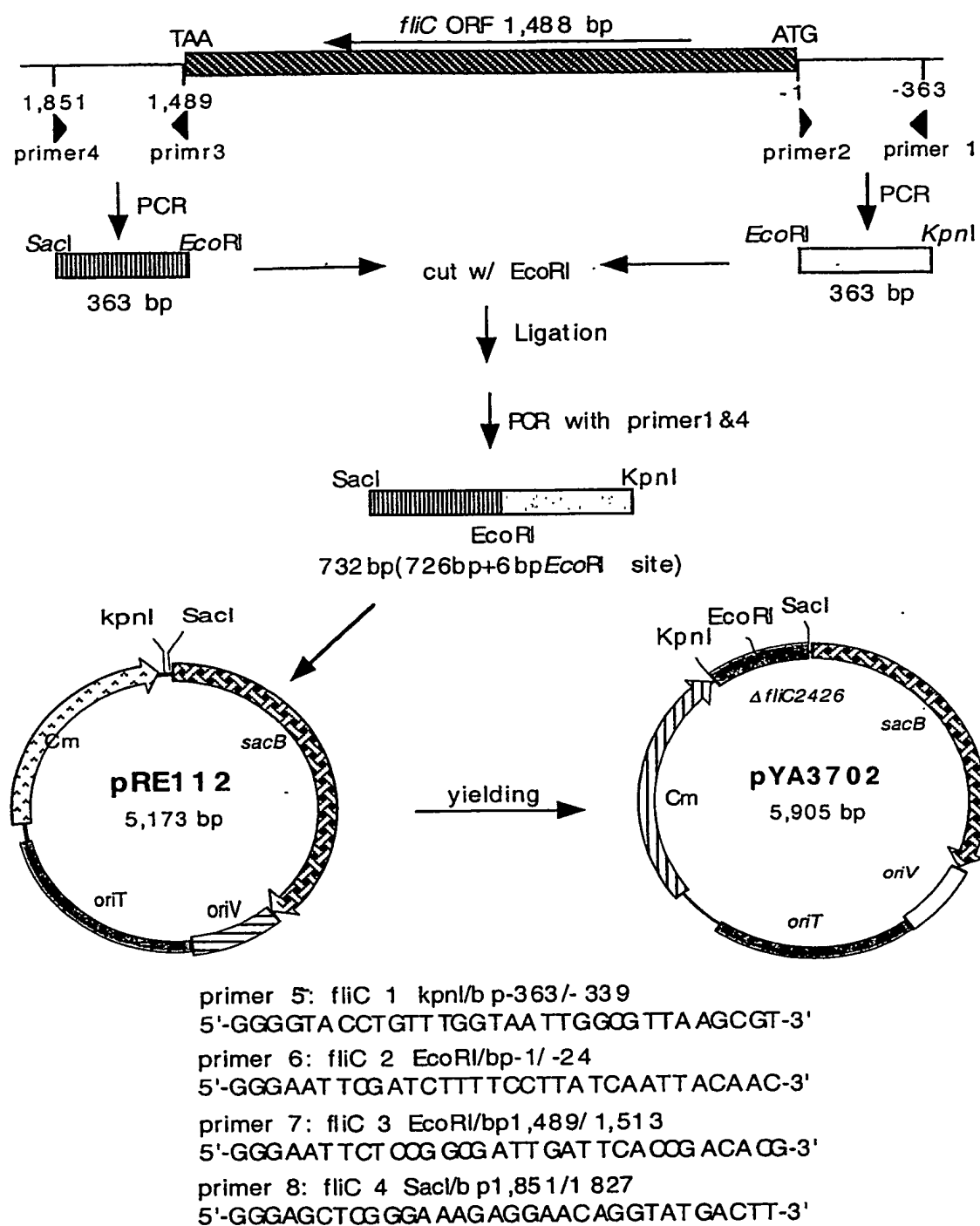
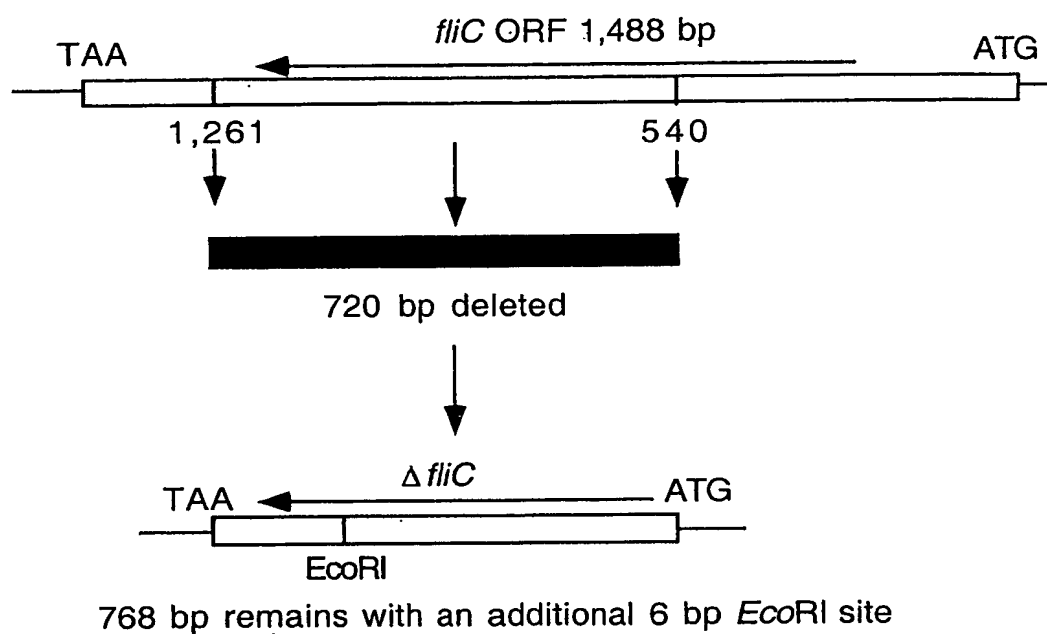


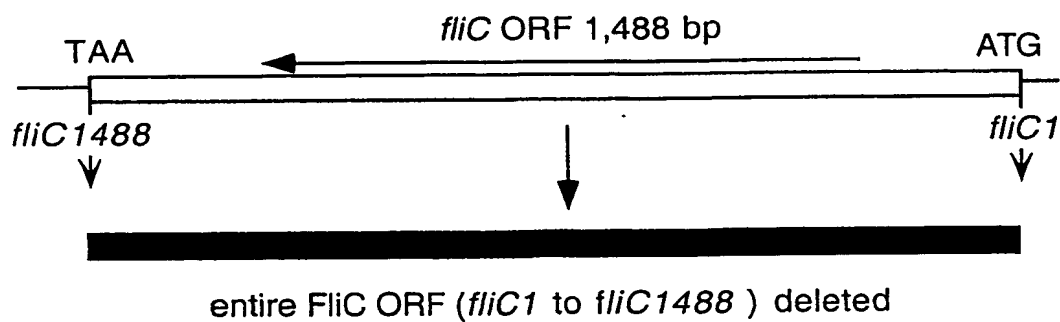
Figure 18. Construction of a suicide vector for transfer of $\Delta fliC$ 2426 mutation

Figure 19. *S. typhimurium* UK-1 chromosomal map for $\Delta fliC$ -Var and $\Delta fliC2426$ deletion mutations.

A. $\Delta fliC$ -Var



B. $\Delta fliC2426$



*araI*₂

5' CCAA AAA AAC GGG TAT GGA GAA ACA GTA GAG AGT TGC GAT AAA AAG CGT CAG GTA GGA 3'

3' GGTT TTT TTG CCC ATA CCT CTT TGT CAT CTC TCA ACG CTA TTT TTC GCA GTC CAT CCT 5'

← *araBAD mRNA* +1 -10 -35

																<i>araI</i> ₁								CRP binding site											
TCC	GCT	AAT	CTT	ATG	GAT	AAA	AAT	GCT	ATG	GCA	TAG	CAA	AGT	GTG	ACG	CCG	TGC	AAA	TAA																
AGG	CGA	TTA	GAA	TAC	CTA	TTT	TTA	CGA	TAC	CGT	ATC	GTT	TCA	CAC	TGC	GGC	ACG	TTT	ATT																

										<i>araO</i> _{1L}				-35				<i>araO</i> _{1R}				-10			
TCA	ATG	TGG	ACT	TTT	CTG	CCG	TGA	TTA	TAG	ACA	CTT	TTG	TTA	CGC	GTT	TTT	GTC	ATG	GCT						
AGT	TAC	ACC	TGA	AAA	GAC	GGC	ACT	AAT	ATC	TGT	GAA	AAC	AAT	GCG	CAA	AAA	CAG	TAC	CGA						

+1 *araC** mRNA →

TTG	GTC	CCG	CTT	TGT	TAC	AGA	ATG	CTT	TTA	ATA	AGC	GGG	GTT	ACC	GGT	TGG	GTT	AGC	GAG
AAC	CAG	GGC	GAA	ACA	ATG	TCT	TAC	GAA	AAT	TAT	TCG	CCC	CAA	TGG	CCA	ACC	CAA	TCG	CTC

																			<i>araO</i> ₂						
AAG	AGC	CAG	TAA	AAG	ACG	CAG	TGA	CGG	CAA	TGT	CTG	ATG	CAA	TAT	GGA	CAA	TTG	GTT	TCT						
TTC	TCG	GTC	ATT	TTC	TGC	GTC	ACT	GCC	GTT	ACA	GAC	TAC	GTT	ATA	CCT	GTT	AAC	CAA	AGA						

↓ *araC** starts

TCT	CTG	AAT	GGT	GGG	AGT	ATG	AAA	AGT	ATG	GCT	GAA	GCG	CAA	AAT	GAT	CCC	CTG	CTG	CCG
									M	A	E	A	Q	N	D	P	L	L	P

GGA	TAC	TCG	TTT	AAC	GCC	CAT	CTG	GTG	GCG	GGT	TTA	ACG	CCG	ATT	GAG	GCC	AAC	GGT	TAT
G	Y	S	F	N	A	H	L	V	A	G	L	T	P	I	E	A	N	G	Y

CTC	GAT	TTT	TTT	ATC	GAC	CGA	CCG	CTG	GGA	ATG	AAA	GGT	TAT	ATT	CTC	AAT	CTC	ACC	ATT
L	D	F	F	I	D	R	P	L	G	M	K	G	Y	I	L	N	L	T	I

CGC	GGT	CAG	GGG	GTG	GTG	AAA	AAT	CAG	GGA	CGA	GAA	TTT	GTC	TGC	CGA	CCG	GGT	GAT	ATT
R	G	Q	G	V	V	K	N	Q	G	R	E	F	V	C	R	P	G	D	I

TTG	CTG	TTC	CCG	CCA	GGA	GAG	ATT	CAT	CAC	TAC	GGT	CGT	CAT	CCG	GAG	GCT	CGC	GAA	TGG
L	L	F	P	P	G	E	I	H	H	Y	G	R	H	P	E	A	R	E	W

TAT	CAC	CAG	TGG	GTT	TAC	TTT	CGT	CCG	CGC	GCC	TAC	TGG	CAT	GAA	TGG	CTT	AAC	TGG	CCG
Y	H	Q	W	V	Y	F	R	P	R	A	Y	W	H	E	W	L	N	W	P

TCA	ATA	TTT	GCC	AAT	ACG	GGT	TTC	TTT	CGC	CCG	GAT	GAA	GCG	CAC	CAG	CCG	CAT	TTC	AGC
S	I	F	A	N	T	G	F	F	R	P	D	E	A	H	Q	P	H	F	S

GAC	CTG	TTT	GGG	CAA	ATC	ATT	AAC	GCC	GGG	CAA	GGG	GAA	GGG	CGC	TAT	TCG	GAG	CTG	CTG
D	L	F	G	Q	I	I	N	A	G	Q	G	E	G	R	Y	S	E	L	L

GCG	ATA	AAT	CTG	CTT	GAG	CAA	TTG	TTA	CTG	CGG	CGC	ATG	GAA	GCG	ATT	AAC	GAG	TCG	CTC
A	I	N	L	L	E	Q	L	L	L	R	R	M							

Figure 20. (cont'd)

TCA CAT CTT TTC CGC CAG CAG TTA GGG ATT AGC GTC TTA AGC TGG CGC GAG GAC CAA CGC
S H L F R Q Q L G I S V L S W R E D Q R

ATT AGT CAG GCG AAG CTG CTT TTG AGC ACT ACC CGG ATG CCT ATC GCC ACC GTC GGT CGC
I S Q A K L L L S T T R M P I A T V G R

AAT GTT GGT TTT GAC GAT CAA CTC TAT TTC TCG CGA GTA TTT AAA AAA TGC ACC GGG GCC
N V G F D D Q L Y F S R V F K K C T G A

AGC CCG AGC GAG TTT CGT GCC GGT TGT GAA GAA AAA GTG AAT GAT GTA GCC GTC AAG TTG
S P S E F R A G C E E K V N D V A V K L

TCA TAA TTG GTA ACG AAT CAG ACA ATT GAC GGC
S *
←araC* ends

Figure 21. DNA and amino acid sequences of *P_{fur}* and *fur* gene of *S. paratyphi* A.*fldA*

181/61 primer 211/71
 GAA GCG CAA TGT GAC TGG GAT GAC TTC TTC CCG ACT CTC GAA GAG ATT GAC TTT AAC GGT
 E A Q C D W D D F F P T L E E I D F N G
 241/81 271/91
 AAG CTG GTG GCG CTG TTT GGC TGT GGC GAT CAG GAA GAC TAC GCG GAA TAC TTC TGT GAT
 K L V A L F G C G D Q E D Y A E Y F C D
 301/101 331/111
 GCG CTG GGC ACG ATT CGC GAC ATT ATT GAG CCG CGC GGC GCC ACG ATT GTG GGT CAC TGG
 A L G T I R D I I E P R G A T I V G H W
 361/121 391/131
 CCA ACT GCA GGC TAT CAT TTT GAA GCC TCT AAA GGT CTG GCT GAC GAC GAT CAT TTT GTC
 P T A G Y H F E A S K G L A D D D H F V
 421/141 451/151
 GGT CTG GCG ATT GAC GAA GAC CGT CAG CCT GAA CTG ACC GCC GAG CGT GTT GAA AAA TGG
 G L A I D E D R Q P E L T A E R V E K W
 481/161 511/171
 GTT AAG CAA GTT TCG GCT GAA TTG CAC CTC GAC GAC ATC CTC AAC GCC TAA TCT TAT GCG
 V K Q V S A E L H L D D I L N A * ↑ *fldA* ends
 541/181 571/191
 GCG CAG CGT TAT ATC TGC GCC GCA TCA ATA GAC AAG ACC AAT CAA AAT AAT TGC TAC AAA
 primer L delete (*fur*-253) OxyR binding site
 601/201 631/211
TTT GTA ACT TTC GCA CCC ATC CCT GTA CAA TGT CCG GGT GTA ATC AGG TGG CGC CAG AAT
 661/221 691/231 -35
TTG CAG GCA AAA CCA CAG TTT TAT TAA CAT CTG CGA GAG ACT TGC GGT TTT CAT TTC GGC
 CRP binding site
 721/241 -10 751/251
 ATG GCA GTC CTA TAA TGA TAC GCA TTA TCT TGA GTG CAA TTT CTG TCA CTT CTC TAA TGA
 Fur consensus
 781/261 SD 813/1
 AGT GAA TCG TTT AGC AAC AGG ACA GAT TCC GC ATG ACT GAC AAC AAT ACC GCA TTA AAG
 delete (*fur*-15) primer M T D N N T A L K
 fur starts ↑
 840/10 873/21
 AAG GCT GGC CTG AAA GTA ACG CTT CCT CGT TTA AAA ATT CTG GAA GTT CTT CAG GAA CCA
 K A G L K V T L P R L K I L E V L Q E P
 900/30 933/41
 GAT AAC CAT CAC GTC AGT GCG GAA GAT TTA TAC AAA CGC CTG ATC GAC ATG GGT GAA GAA
 D N H H V S A E D L Y K R L I D M G E E
 960/50 993/61
 ATC GGT CTG GCA ACC GTA TAC CGT GTG CTG AAC CAG TTT GAC GAT GCC GGT ATC GTG ACC
 I G L A T V Y R V L N Q F D D A G I V T
 1020/70 1053/81
 CGC CAT AAT TTT GAA GGC GGT AAA TCC GTT TTT GAA CTG ACG CAA CAG CAT CAT CAC GAC
 R H N F E G G K S V F E L T Q Q H H H D
 1080/90 primer 1113/101
 CAT CTT ATC TGC CTT GAT TGC GGA AAA GTG ATT GAA TTT AGT GAT GAC TCT ATT GAA GCG
 H L I C L D C G K V I E F S D D S I E A
 1140/110 1173/121
 CGC CAG CGT GAA ATT GCG GCG AAA CAC GGT ATT CGT TTA ACT AAT CAC AGC CTC TAT CTT
 R Q R E I A A K H G I R L T N H S L Y L
 1200/130 1233/141
 TAC GGC CAC TGC GCT GAA GGC GAC TGC CGC GAA GAC GAG CAC GCG CAC GAT GAC GCG ACT
 Y G H C A E G D C R E D E H A H D D A T
 1260/150
 AAA TAA
 K * *fur* ends

Figur 22. (cont'd)

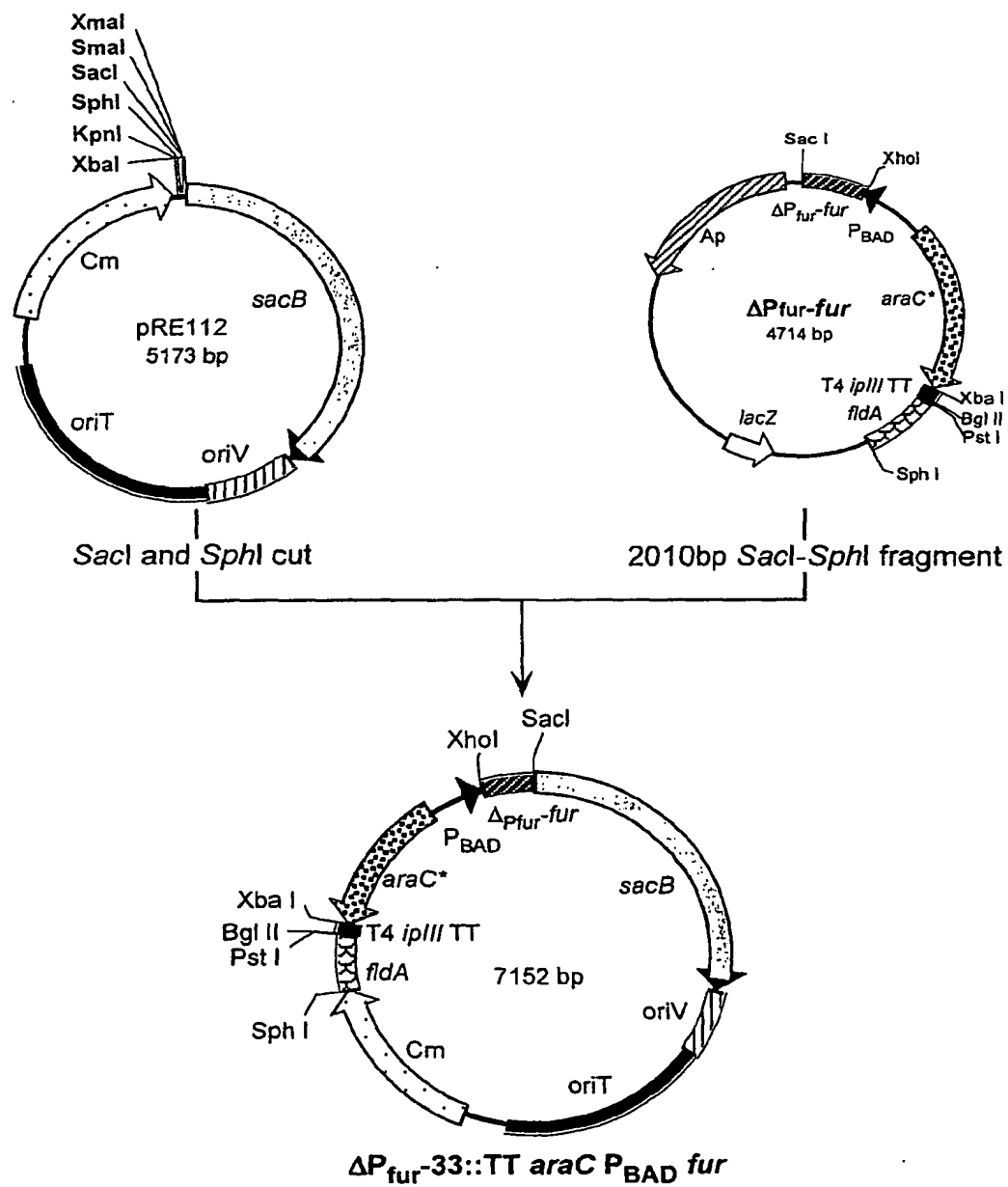
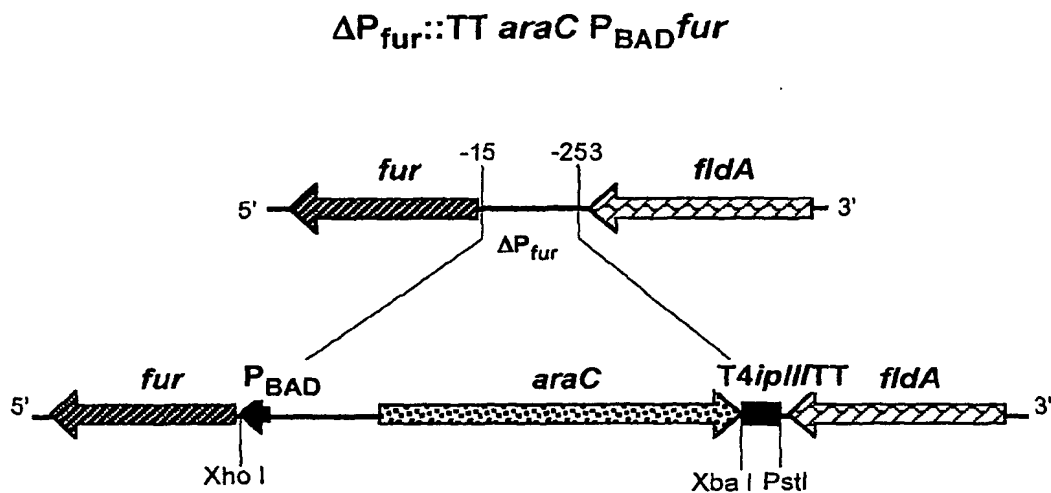


Figure 23. Chromosomal map of ΔP_{fur} -33::TT *araC* P_{BAD} *fur* deletion-insertion mutation.



fur promoter region (-15 to -253; including Fur consensus, CRP binding, and OxyR binding sites) deleted and 1344 bp P_{BAD} *araC* TT inserted.

Figure 24. DNA sequence of the $\Delta P_{fur-33}::TT\ araC^* P_{BAD}\ fur$.*fldA*

R Q P E L T A E R V E K W V K Q V S A E
CGT CAG CCT GAA CTG ACC GCC GAG CGT GTT GAA AAA TGG GTT AAG CAA GTT TCG GCT GAA

L H L D D I L N A * S Y A A Q R Y I C A
TTG CAC CTC GAC GAC ATC CTC AAC GCC TAA TCT TAT GCG GCG CAG CGT TAT ATC TGC GCT

←*fldA* ends*fur*₋₂₅₄*Pst*I

GCA GAG ATC TTT TAT TAT TCT ATC CTA GAA TTG TGA TAA TAT ATT CAC AAT TCT AGG AGT
T4 *ipIII* transcription terminator sequence

*Xba*I

TGT AAA CTG CTT TTA TTT ATC TAG AGT CAA GCC GTC AAT TGT CTG ATT CGT TAC CAA TTA
ACA TTT GAC GAA AAT AAA TAG ATC TCA GTT CGG CAG TTA ACA GAC TAA GCA ATG GTT AAT

araC ends →

TGA CAA CTT GAC GGC TAC ATC ATT CAC TTT TTC TTC ACA ACC GGC ACG GAA CTC GCT CGG
ACT GTT GAA CTG CCG ATG TAG TAA GTG AAA AAG AAG TGT TGG CCG TGC CTT GAG CGA GCC
S L K V A V D N V K E E C G A R F E S P

GCT GGC CCC GGT GCA TTT TTT AAA TAC CCG CGA GAA ATA GAG TTG ATC GTC AAA ACC AAC
CGA CCG GGG CCA CGT AAA AAA TTT ATG GGC GCT CTT TAT CTC AAC TAG CAG TTT TGG TTG
S A G T C K K F V R S F Y L Q D D F G V

ATT GCG ACC GAC GGT GGC GAT AGG CAT CCG GGT GGT GCT CAA AAG CAG CTT CGC CTG GCT
TAA CGC TGG CTG CCA CCG CTA TCC GTA GGC CCA CCA CGA GTT TTC GTC GAA GCG GAC CGA
N R G V T A I P M R T T S L L L K A Q S

GAT ACG TTG GTC CTC GCG CCA GCT TAA GAC GCT AAT CCC TAA CTG CTG GCG GAA AAG ATG
CTA TGC AAC CAG GAG CGC GGT CGA ATT CTG CGA TTA GGG ATT GAC GAC CGC CTT TTC TAC
I R Q D E R W S L V S I G L Q Q R F L H

TGA CAG ACG CGA CCG CGA CAA GCA AAC ATG CTG TGC GAC GCT GGC GAT ATC AAA ATT GCT
ACT GTC TGC GCT GCC GCT GTT CGT TTG TAC GAC ACG CTG CGA CCG CTA TAG TTT TAA CGA
S L R S P S L C V H Q A V S A I D F N S

GTC TGC CAG GTG ATC GCT GAT GTA CTG ACA AGC CTC GCG TAC CCG ATT ATC CAT CGG TGG
CAG ACG GTC CAC TAG CGA CTA CAT GAC TGT TCG GAG CGC ATG GGC TAA TAG GTA GCC ACC
D A L H D S I Y Q C A E R V R N D M P P

ATG GAG CGA CTC GTT AAT CGC TTC CAT GCG CCG CAG TAA CAA TTG CTC AAG CAG ATT TAT
TAC CTC GCT GAG CAA TTA GCG AAG GTA CGC GGC GTC ATT GTT AAC GAG TTC GTC TAA ATA
H L S E N I A E M R R L L L Q E L L N I

CGC CAG CAG CTC CGA ATA GCG CCC TTC CCC TTG CCC GGC GTT AAT GAT TTG CCC AAA CAG
GCG GTC GTC GAG GCT TAT CGC GGG AAG GGG AAC GGG CCG CAA TTA CTA AAC GGG TTT GTC
A L L E S Y R G E G Q G A N I I Q G F L

GTC GCT GAA ATG CGG CTG GTG CGC TTC ATC CGG GCG AAA GAA CCC CGT ATT GGC AAA TAT
CAG CGA CTT TAC GCC GAC CAC GCG AAG TAG GCC CGC TTT CTT GGG GCA TAA CCG TTT ATA
D S F H P Q H A E D P R F F G T N A F I

TGA CGG CCA GTT AAG CCA TTC ATG CCA GTA GGC GCG CGG ACG AAA GTA AAC CCA CTG GTG
ACT GCC GGT CAA TTC GGT AAG TAC GGT CAT CCG CGC GCC TGC TTT CAT TTG GGT GAC CAC
S P W N L W E H W Y A R P R F Y V W Q H

Figure 24. (cont'd)

ATA CCA TTC GCG AGC CTC CGG ATG ACG ACC GTA GTG ATG AAT CTC TCC TGG CGG GAA CAG
 TAT GGT AAG CGC TCG GAG GCC TAC TGC TGG CAT CAC TAC TTA GAG AGG ACC GCC CTT GTC
 Y W E R A E P H R G Y H H I E G P P F L

 CAA AAT ATC ACC CGG TCG GCA AAC AAA TTC TCG TCC CTG ATT TTT CAC CAC CCC CTG ACC
 GTT TTA TAG TGG GCC AGC CGT TTG TTT AAG AGC AGG GAC TAA AAA GTG GTG GGG GAC TGG
 L I D G P R C V F E R G Q N K V V G Q G

 GCG AAT GGT GAG ATT GAG AAT ATA ACC TTT CAT TCC CAG CGG TCG GTC GAT AAA AAA ATC
 CGC TTA CCA CTC TAA CTC TTA TAT TGG AAA GTA AGG GTC GCC AGC CAG CTA TTT TTT TAG
 R I T L N L I Y G K M G L P R D I F F D

 GAG ATA ACC GTT GGC CTC AAT CGG CGT TAA ACC CGC CAC CAG ATG GGC ATT AAA CGA GTA
 CTC TAT TGG CAA CCG GAG TTA GCC GCA ATT TGG GCG GTG GTC TAC CCG TAA TTT GCT CAT
 L Y G N A E I P T L G A V L H A N F S Y

 TCC CGG CAG CAG GGG ATC ATT TTG CGC TTC AGC CAT ACT TTT CAT ACT CCC GCC ATT CAG
 AGG GCC GTC GTC CCC TAG TAA AAC GCG AAG TCG GTA TGA AAA GTA TGA GGG CGG TAA GTC
 G P L L P D N Q A E A M
 ← araC starts

 AGA AGA AAC CAA TTG TCC ATA TTG CAT CAG ACA TTG CCG TCA CTG CGT CTT TTA CTG GCT
 TCT TCT TTG GTT AAC AGG TAT AAC GTA GTC TGT AAC GGC AGT GAC GCA GAA AAT GAC CGA
 araO₂

 CTT CTC GCT AAC CAA ACC GGT AAC CCC GCT TAT TAA AAG CAT TCT GTA ACA AAG CGG GAC
 GAA GAG CGA TTG GTT TGG CCA TTG GGG CGA ATA ATT TTC GTA AGA CAT TGT TTC GCC CTG

 CAA AGC CAT GAC AAA AAC GCG TAA CAA AAG TGT CTA TAA TCA CGG CAG AAA AGT CCA CAT
 GTT TCG GTA CTG TTT TTG CGC ATT GTT TTC ACA GAT ATT AGT GCC GTC TTT TCA GGT GTA
 +laraC mRNA -10 araO_{1R} -35 araO_{1L}

 TGA TTA TTT GCA CGG CGT CAC ACT TTG CTA TGC CAT AGC ATT TTT ATC CAT AAG ATT AGC
 ACT AAT AAA CGT GCC GCA GTG TGA AAC GAT ACG GTA TCG TAA AAA TAG GTA TTC TAA TCG
 CRP binding site araI₁
 -35 -10 +1 araBAD mRNA
 GGA TCC TAC CTG ACG CTT TTT ATC GCA ACT CTC TAC TGT TTC TCC ATA CCC GTT TTT TTG
 CCT AGG ATG GAC TGC GAA AAA TAG CGT TGA GAG ATG ACA AAG AGG TAT GGG CAA AAA AAC
 araI₂ fur starts→
 xhoI ↓fur₋₁₄ M T D N N T A L K K A
 GGC TAG CCT CGA GAG GAC AGA TTC CGC ATG ACT GAC AAC AAT ACC GCA TTA AAG AAG GCT
 CCG ATC GGA GCT CTC CTG TCT AAG GCG TAC TGA CTG TTG TTA TGG CGT AAT TTC TTC CGA

 G L K V T L P R L K I L E V L Q E P D N
 GGC CTG AAA GTA ACG CTT CCT CGT TTA AAA ATT CTG GAA GTT CTT CAG GAA CCA GAT AAC
 CCG GAC TTT CAT TGC GAA GGA GCA AAT TTT TAA GAC CTT CAA GAA GTC CTT GGT CTA TTG

 H H V S A E D L Y K R L I D M G E E I G
 CAT CAC GTC AGT GCG GAA GAT TTA TAC AAA CGC CTG ATC GAC ATG GGT GAA GAA ATC GGT
 GTA GTG CAG TCA CGC CTT CTA AAT ATG TTT GCG GAC TAG CTG TAC CCA CTT CTT TAG CCA

 L A T V Y R V L N Q F D D A G I V T R H
 CTG GCA ACC GTA TAC CGT GTG CTG AAC CAG TTT GAC GAT GCC GGT ATC GTG ACC CGC CAT
 GAC CGT TGG CAT ATG GCA CAC GAC TTG GTC AAA CTG CTA CGG CCA TAG CAC TGG GCG GTA

Figure 24. (cont'd)

N F E G G K S V F E L T Q Q H H H D H L
 AAT TTT GAA GGC GGT AAA TCC GTT TTT GAA CTG ACG CAA CAG CAT CAT CAC GAC CAT CTT
 TTA AAA CTT CCG CCA TTT AGG CAA AAA CTT GAC TGC GTT GTC GTA GTA GTG CTG GTA GAA

I C L D C G K V I E F S D D S I E A R Q
 ATC TGC CTT GAT TGC GGA AAA GTG ATT GAA TTT AGT GAT GAC TCT ATT GAA GCG CGC CAG
 TAG ACG GAA CTA ACG CCT TTT CAC TAA CTT AAA TCA CTA CTG AGA TAA CTT CGC GCG GTC

R E I A A K H G I R L T N H S L Y L Y G
 CGT GAA ATT GCG GCG AAA CAC GGT ATT CGT TTA ACT AAT CAC AGC CTC TAT CTT TAC GGC
 GCA CTT TAA CGC CGC TTT GTG CCA TAA GCA AAT TGA TTA GTG TCG GAG ATA GAA ATG CCG

H C A E G D C R E D E H A H D D A T K * ←fur ends
 CAC TGC GCT GAA GGC GAC TGC CGC GAA GAC GAG CAC GCG CAC GAT GAC GCG ACT AAA TAA
 GTG ACG CGA CTT CCG CTG ACG GCG CTT CTG CTC GTG CGC GTG CTA CTG CGC TGA TTT ATT

Figure 25. DNA and amino acid sequences of P_{rpoS} , *rpoS* and flanking region of *S. typhimurium* and *S. typhi*.

STM: *S. typhimurium* 14028S

STY: *S. typhi* CT18

AAT GCA AGC AGT ACG TCA ACC AGC GCG CCG ATT TCC GCA TGG CGC TGG CCG ACG GAT GGC-STM
AAT GCA AGC AGT ACG TCA ACC AGC GCG CCG ATT TCC GCA TGG CGC TGG CCG ACG GAT GGC-STY
N A S S T S T S A P I S A W R W P T D G

AAA GTG ATC GAA AAC TTT GGC GCT TCC GAA GGG GGC AAT AAA GGG ATC GAC ATT GCA GGC
AAA GTG ATC GAA AAC TTT GGC GCT TCC GAA GGG GGC AAT AAA GGG ATC GAC ATT GCA GGC
K V I E N F G A S E G G N K G I D I A G

AGT AAG GGA CAG GCT ATC GTC GCA ACC GCT GAT GGG CGC GTC GTA TAT GCC GGT AAC GCA
AGT AAG GGA CAG GCT ATC GTC GCA ACC GCT GAT GGG CGC GTC GTA TAT GCC GGT AAC GCA
S K G Q A I V A T A D G R V V Y A G N A

CTG CGT GGT TAC GGT AAT CTT ATT ATC ATC AAA CAT AAC GAT GAT TAC CTG AGT GCC TAC
CTG CGT GGT TAC GGT AAT CTT ATT ATC ATC AAA CAT AAC GAT GAT TAC CTG AGT GCC TAC
L R G Y G N L I I I K H N D D Y L S A Y

GCC CAT AAT GAT ACG ATG CTG GTC CGG GAA CAA CAG GAA GTT AAG GCG GGG CAA AAA ATC
GCC CAT AAT GAT ACG ATG CTG GTC CGG GAA CAA CAG GAA GTT AAG GCG GGG CAA AAA ATC
A H N D T M L V R E Q Q E V K A G Q K I

GCT ACT ATG GGT AGC ACC GGC ACC AGC TCT ACA CGC TTG CAT TTT GAA ATT CGT TAC AAG
GCT ACT ATG GGT AGC ACC GGC ACC AGC TCT ACA CGC TTG CAT TTT GAA ATT CGT TAC AAG
A T M G S T G T S S T R L H F E I R Y K

GGG AAA TCC GTA AAC CCG CTG CGT TAT TTA CCG CAG CGA TAA AG
GGG AAA TCC GTA AAC CCG CTG CGT TAT TTA CCG CAG CGA TAA AG
G K S V N P L R Y L P Q R *

← *nlpD* ends

CGG CGG AAC CAG GCT TTG ACT TGC TAG TTC CGT CAA GGG ATC ACG GGT AGG AGC CAC CTT
CGG CGG AAC CAG GCT TTG ACT TGC TAG TTC CGT CAA GGG ATC ACG GGT AGG AGC CAC CTT
↑ ↑

rpoS₋₄₈

ΔP_{rpoS} (*rpoS*-48 to -13 deleted)

rpoS₋₁₃

1185/1

1215/11

ATG AGT CAG AAT ACG CTG AAA GTT CAT GAT TTA AAT GAA GAC GCG GAA TTT GAT GAG AAC-STM
ATG AGT CAG AAT ACG CTG AAA GTT CAT GAT TTA AAT GAA GAC GCG GAA TTT GAT GAG AAC-STY
M S Q N T L K V H D L N E D A E F D E N

rpoS starts →

1245/21

1275/31

GGA GTA GAG GCT TTT GAC GAA AAA GCC TTG AGT GAA GAG GAA CCC AGT GAT AAC GAC CTG
GGA GTA GAG GCT TTT GAC GAA AAA GCC TTG AGT GAA GAG GAA CCC AGT GAT AAC GAC CTG
G V E A F D E K A L S E E E P S D N D L

1305/41

1335/51

GCT GAA GAA GAG CTG TTA TCG CAA GGG GCC ACA CAG CGT GTG TTG GAC GCG ACT CAG CTT
GCT GAA GAA GAG CTG TTA TCG CAA GGG GCC ACA CAG CGT GTG TTG GAC GCG ACT CAG CTT
A E E E L L S Q G A T Q R V L D A T Q L

1365/61

1395/71

TAC CTT GGT GAG ATT GGG TAT TCA CCA CTG TTA ACA GCC GAA GAA GAA GTC TAT TTT GCG
TAC CTT GGT GAG ATT GGG TAT TCA CCA CTG TTA ACA GCC GAA GAA GAA GTC TAT TTT GCG
Y L G E I G Y S P L L T A E E E V Y F A

1425/81

1455/91

CGT CGC GAC CTG CGT GGA GAT GTC GCT TCT CGC CGT CGC ATG ATT GAG AGT AAC CTG CGT
CGT CGC GCA CTG CGT GGA GAT GTC GCT TCT CGC CGT CGC ATG ATT GAG AGT AAC CTG CGT
R R A L R G D V A S R R R M I E S N L R

1485/101

1515/111

CTG GTG GTA AAA ATT GCC CGC CGT TAT GGC AAT CGT GGA CTG GCG TTG CTG GAC CTG ATT
CTG GTG GTA AAA ATT GCC CGC CGT TAT GGC AAT CGT GGA CTG GCG TTG CTG GAC CTG ATT
L V V K I A R R Y G N R G L A L L D L I

Figure 25. (cont'a)

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1545/121
GAA GAG GGC AAC CTG GGG CTT ATC CGT GCA GTA GAG AAG TTT GAC CCG GAA CGC GGG TTC
GAA GAG GGC AAC CTG GGG CTT ATC CGT GCA GTC GAG AAG TTT GAC CCG GAA CGC GGG TTC
E E G N L G L I R A V E K F D P E R G F
1605/141
CGC TTC TCA ACA TAC GCA ACC TGG TGG ATT CGC CAG ACA ATC GAA CGG GCG ATC ATG AAC
CGC TTC TCA ACA TAC GCA ACC TGG TGG ATT CGC CAG ACA ATC GAA CGG GCG ATT ATG AAC
R F S T Y A T W W I R Q T I E R A I M N
1665/161
CAA ACC CGT ACG ATT CGC TTG CCG ATT CAC ATT GTT AAA GAG CTG AAC GTA TAC CTG CGC
CAA ACC CGT ACG ATT CGC TTG CCG ATT CAC ATT GTT AAA GAG CTG AAC GTA TAC CTG CGC
Q T R T I R L P I H I V K E L N V Y L R
1725/181
ACC GCA CGT GAG TTG TCG CAT AAA CTG GAC CAC GAA CCG AGT GCG GAA GAA ATT GCA GAG
ACC GCA CGT GAG TTG TCG CAT AAA CTG GAC CAC GAA CCG AGT GCG GAA GAA ATT GCA GAG
T A R E L S H K L D H E P S A E E I A E
1785/201
CAA CTG GAT AAA CCG GTT GAT GAC GTC AGC CGT ATG CTT CGT CTC AAC GAG CGC ATT ACC
CAA CTG GAT AAA CCG GTT GAT GAC GTC AGC CGT ATG CTT CGT CTC AAC GAG CGC ATT ACC
Q L D K P V D D V S R M L R L N E R I T
1845/221
TCG GTA GAC ACC CCG CTG GGC GGT GAT TCC GAA AAA GCG TTG CTG GAC ATC CTG GCC GAT
TCG GTA GAC ACC CCG CTG GGC GGT GAT TCC GAA AAA GCG TTG CTG GAC ATC CTG GCC GAT
S V D T P L G G D S E K A L L D I L A D
1905/241
GAA AAA GAG AAC GGT CCG GAA GAC ACC ACG CAA GAT GAC GAT ATG AAA CAG AGC ATC GTC
GAA AAA GAG AAC GGT CCG GAA GAC ACC ACG CAA GAT GAC GAT ATG AAA CAG AGC ATC GTC
E K E N G P E D T T Q D D D M K Q S I V
1965/261
AAA TGG TTG TTC GAA CTG AAC GCC AAA CAG CGT GAA GTG CTG GCG CGC CGT TTC GGT CTG
AAA TGG TTG TTC GAA CTG AAC GCC AAA CAG CGT GAA GTG CTG GCG CGC CGT TTC GGT CTG
K W L F E L N A K Q R E V L A R R F G L
2025/281
CTG GGA TAT GAA GCT GCG ACA CTG GAA GAT GTA GGC CGT GAA ATC GGT CTT ACG CGT GAA
CTG GGA TAT GAA GCT GCG ACA CTG GAA GAT GTA GGC CGT GAA ATC GGT CTT ACG CGT GAA
L G Y E A A T L E D V G R E I G L T R E
2085/301
CGT GTT CGT CAG ATT CAG GTT GAA GGC CTG CGC CGT CTG CGC GAA ATT CTG CAG ACG CAG
CGT GTT CGT CAG ATT CAG GTT GAA GGC CTG CGC CGT CTG CGC GAA ATT CTG CAG ACG CAG
R V R Q I Q V E G L R R L R E I L Q T Q
2145/321
GGG CTG AAT ATC GAA GCG CTG TTC CGC GAG TAA GTA CCC TTG TCA
GGG CTG AAT ATC GAA GCG CTG TTC CGC GAG TAA GTA CCC TTG TCA
G L N I E A L F R E *
← rpoS ends

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Figure 26. Construction of suicide vector for introducing ΔP_{rpoS} -183::TT *araC* P_{BAD} *rpoS* deletion-insertion mutation.

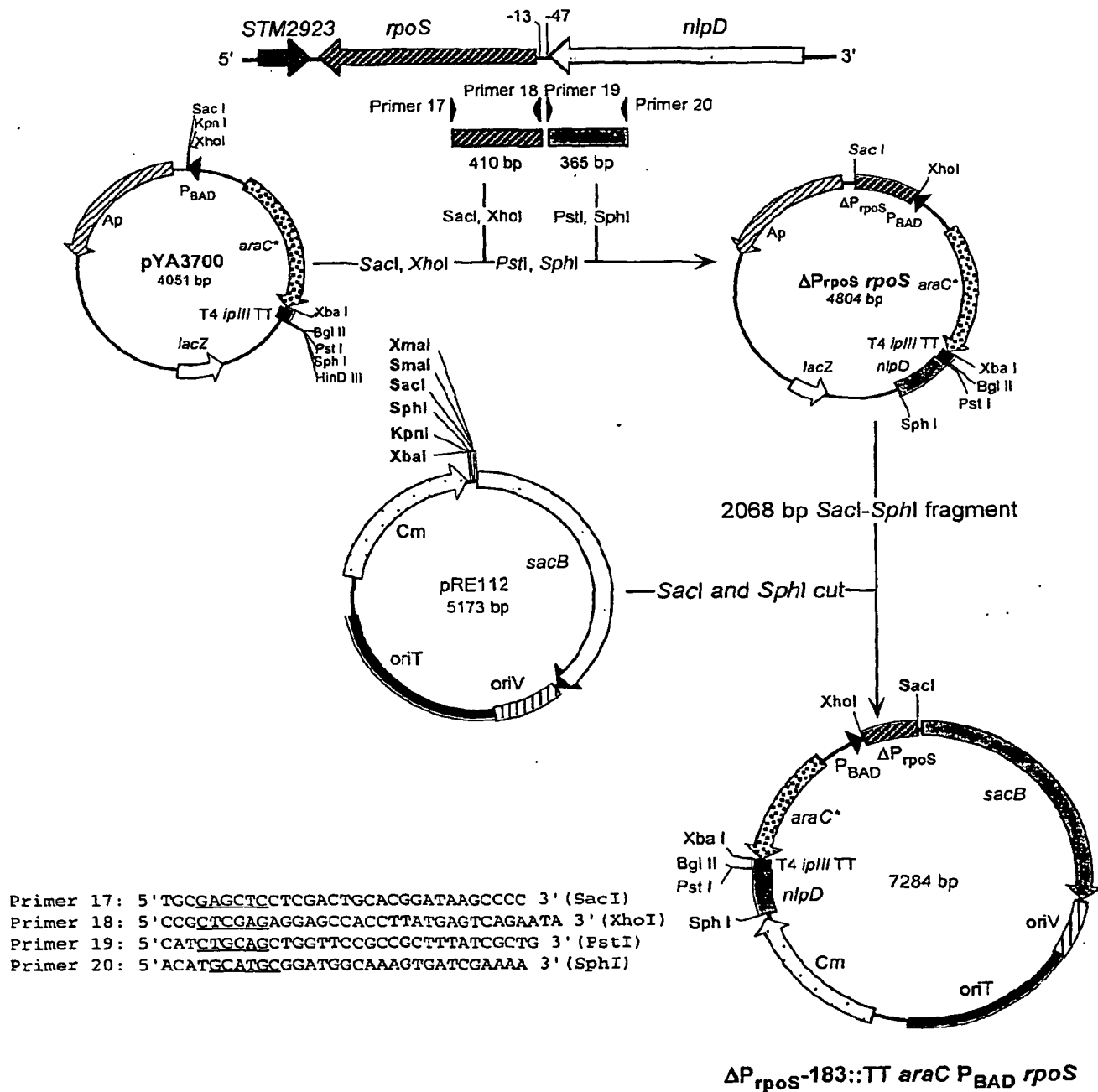
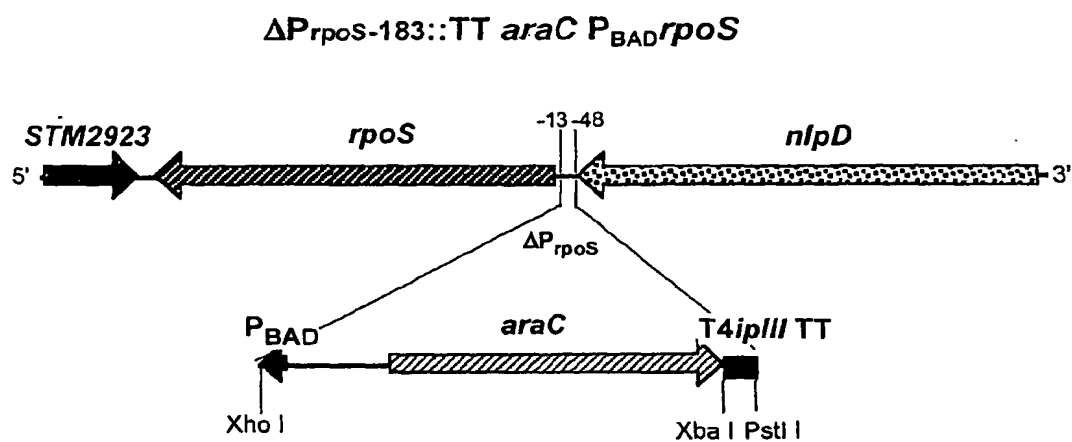


Figure 27. Chromosomal map of $\Delta P_{rpoS-183::TT} \text{ araC } P_{BAD} rpoS$ deletion-insertion mutation.



rpoS promoter region (-13 to -48) deleted and 1344 bp $P_{BAD} \text{ araC } TT$ inserted.

Figure 28. DNA and amino acid sequences of the *S. typhimurium* *P_{phoPQ}* and *phoPQ* and the flanking region.

***PurB/asl* →**

ATT GCG TTG AAC CAT TTC AAA CAG AAA ACC ATC GCC GGG GAG ATC GGT TCT TCT ACC ATG
I A L N H F K Q K T I A G E I G S S T M

CCG CAT AAA GTT AAC CCC ATT GAC TTT GAA AAC TCA GAA GGC AAC CTC GGT CTG TCT AAC
P H K V N P I D F E N S E G N L G L S N

GCA GTG TTG CAC CAT CTG GCA AAC AAA CTG CCG GTT TCC CGC TGG CAG CGC GAT CTG ACC
A V L H H L A N K L P V S R W Q R D L T

GAC TCA ACC GTC CTG CGT AAC CTG GGT GTC GGC ATC GGC TAT GCG CTT ATC GCT TAT CAG
D S T V L R N L G V G I G Y A L I A Y Q

TCC ACC CTG AAG GGC GTC AGC AAG CTG GAA GTA AAC CGC GAT CAT CTG CTT GAC GAA CTG
S T L K G V S K L E V N R D H L L D E L

GAT CAC AAC TGG GAA GTA TTA GCC GAA CCG ATC CAG ACC GTC ATG CGC CGC TAT GGT ATT
D H N W E V L A E P I Q T V M R R Y G I

GAA AAA CCA TAT GAA AAA CTG AAA GAG TTG ACC CGT GGC AAG CGT GTT GAT GCC GAA GGA
E K P Y E K L K E L T R G K R V D A E G

ATG AAA CAG TTT ATT GAT AGT CTG GCC CTG CCG GAA GCA GAA AAA ACG CGC CTT AAA GCC
M K Q F I D S L A L P E A E K T R L K A

ATG ACG CCG GCA AAT TAT ATC GGT CGC GCT GTG ACT CTG GTC GAC GAA CTT AAA TAA TGC
M T P A N Y I G R A V T L V D E L K *

←*purB* ends

CTG CCT CAC CCT CTT TTC TTC AGA AAG AGG GTG ACT ATT TGT CTG GTT TAT TAA CTG TTT
↑
phoPQ₋₁₀₉

ΔP_{phoPQ} (*phoPQ*-109 to *phoPQ*-12 deleted)

ATC CCC AAA GCA CCA TAA TCA ACG CTA GAC TGT TCT TAT TGT TAA CAC AAG GGA GAA GAG
↑ SD

phoPQ₋₁₂

723/1

ATG ATG CGC GTA CTG GTT GTA GAG GAT AAT GCA TTA TTA CGC CAC CAC CTG AAG GTT CAG
M M R V L V V E D N A L L R H H L K V Q

phoP starts →

753/11

783/21

CTC CAG GAT TCA GGT CAC CAG GTC GAT GCC GCA GAA GAT GCC AGG GAA GCT GAT TAC TAC
L Q D S G H Q V D A A E D A R E A D Y Y

843/41

CTT AAT GAA CAC CTT CCG GAT ATC GCT ATT GTC GAT TTA GGT CTG CCG GAT GAA GAC GGC
L N E H L P D I A I V D L G L P D E D G

903/61

CTT TCC TTA ATA CGC CGC TGG CGC AGC AGT GAT GTT TCA CTG CCG GTT CTG GTG TTA ACC
L S L I R R W R S S D V S L P V L V L T

963/81

GCG CGC GAA GGC TGG CAG GAT AAA GTC GAG GTT CTC AGC TCC GGG GCC GAT GAC TAC GTG
A R E G W Q D K V E V L S S G A D D Y V

1023/101

ACG AAG CCA TTC CAC ATC GAA GAG GTA ATG GCG CGT ATG CAG GCG TTA ATG CGC CGT AAT
T K P F H I E E V M A R M Q A L M R R N

1053/111

Figure 28. (cont'd)

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1083/121      1113/131
AGC GGT CTG GCC TCC CAG GTG ATC AAC ATC CCG CCG TTC CAG GTG GAT CTC TCA CGC CGG
S   G   L   A   S   Q   V   I   N   I   P   P   F   Q   V   D   L   S   R   R
1143/141      1173/151
GAA TTA TCC GTC AAT GAA GAG GTC ATC AAA CTC ACG GCG TTC GAA TAC ACC ATT ATG GAA
E   L   S   V   N   E   E   V   I   K   L   T   A   F   E   Y   T   I   M   E
1203/161      1233/171
ACG CTT ATC CGT AAC AAC GGT AAA GTG GTC AGC AAA GAT TCG CTG ATG CTT CAG CTG TAT
T   L   I   R   N   N   G   K   V   V   S   K   D   S   L   M   L   Q   L   Y
1263/181      1293/191
CCG GAT GCG GAA CTG CGG GAA AGT CAT ACC ATT GAT GTT CTC ATG GGG CGT CTG CGG AAA
P   D   A   E   L   R   E   S   H   T   I   D   V   L   M   G   R   L   R   K
1323/201      1353/211
AAA ATA CAG GCC CAG TAT CCG CAC GAT GTC ATT ACC ACC GTA CGC GGA CAA GGA TAT CTT
K   I   Q   A   Q   Y   P   H   D   V   I   T   T   V   R   G   Q   G   Y   L

1383/221      ← phoP ends
TTT GAA TTG CGC TAA TGA
F   E   L   R   *   *

                phoQ starts →      1415/11
                ATG AAT AAA TTT GCT CGC CAT TTT CTG CCG CTG TCG CTG CGG GTT CGT
                M   N   K   F   A   R   H   F   L   P   L   S   L   R   V   R

1445/21      1475/31
TTT TTG CTG GCG ACA GCC GGC GTC GTG CTG GTG CTT TCT TTG GCA TAT GGC ATA GTG GCG
F   L   L   A   T   A   G   V   V   L   V   L   S   L   A   Y   G   I   V   A
1505/41      1535/51
CTG GTC GGC TAT AGC GTA AGT TTT GAT AAA ACC ACC TTT CGT TTG CTG CGC GGC GAA AGC
L   V   G   Y   S   V   S   F   D   K   T   T   F   R   L   L   R   G   E   S
1565/61      1595/71
AAC CTG TTT TAT ACC CTC GCC AAA TGG GAA AAT AAT AAA ATC AGC GTT GAG CTG CCT GAA
N   L   F   Y   T   L   A   K   W   E   N   N   K   I   S   V   E   L   P   E
1625/81      1655/91
AAT CTG GAC ATG CAA AGC CCG ACC ATG ACG CTG ATT TAC GAT GAA ACG GGC AAA TTA TTA
N   L   D   M   Q   S   P   T   M   T   L   I   Y   D   E   T   G   K   L   L
1685/101      1715/111
TGG ACG CAG CGC AAC ATT CCC TGG CTG ATT AAA AGC ATT CAA CCG GAA TGG TTA AAA ACG
W   T   Q   R   N   I   P   W   L   I   K   S   I   Q   P   E   W   L   K   T
1745/121      1775/131
AAC GGC TTC CAT GAA ATT GAA ACC AAC GTA GAC GCC ACC AGC ACG CTG TTG AGC GAA GAC
N   G   F   H   E   I   E   T   N   V   D   A   T   S   T   L   L   S   E   D
1805/141      1835/151
CAT TCC GCG CAG GAA AAA CTC AAA GAA GTA CGT GAA GAT GAC GAT GAT GCC GAG ATG ACC
H   S   A   Q   E   K   L   K   E   V   R   E   D   D   D   D   D   A   E   M   T
1865/161      1895/171
CAC TCG GTA GCG GTA AAT ATT TAT CCT GCC ACG GCG CGG ATG CCG CAG TTA ACC ATC GTG
H   S   V   A   V   N   I   Y   P   A   T   A   R   M   P   Q   L   T   I   V
1925/181      1955/191
GTG GTC GAT ACC ATT CCG ATA GAA CTA AAA CGC TCC TAT ATG GTG TGG AGC TGG TTC GTA
V   V   D   T   I   P   I   E   L   K   R   S   Y   M   V   W   S   W   F   V
1985/201      2015/211
TAC GTG CTG GCC GCC AAT TTA CTG TTA GTC ATT CCT TTA CTG TGG ATC GCC GCC TGG TGG
Y   V   L   A   A   N   L   L   L   V   I   P   L   L   W   I   A   A   W   W
2045/221      2075/231
AGC TTA CGC CCT ATC GAG GCG CTG GCG CGG GAA GTC CGC GAG CTT GAA GAT CAT CAC CGC
S   L   R   P   I   E   A   L   A   R   E   V   R   E   L   E   D   H   H   R
2105/241      2135/251
GAA ATG CTC AAT CCG GAG ACG ACG CGT GAG CTG ACC AGC CTT GTG CGC AAC CTT AAT CAA
E   M   L   N   P   E   T   T   R   E   L   T   S   L   V   R   N   L   N   Q
2165/261      2195/271

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Figure 28. (cont'd)

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CTG CTC AAA AGC GAG CGT GAA CGT TAT AAC AAA TAC CGC ACG ACC CTG ACC GAC CTG ACG
L  L  K  S  E  R  E  R  Y  N  K  Y  R  T  T  L  T  D  L  T
2225/281
CAC AGT TTA AAA ACG CCG CTC GCG GTT TTG CAG AGT ACG TTA CGC TCT TTA CGC AAC GAA
H  S  L  K  T  P  L  A  V  L  Q  S  T  L  R  S  L  R  N  E
2285/301
AAG ATG AGC GTC AGC AAA GCT GAA CCG GTG ATG CTG GAA CAG ATC AGC CGG ATT TCC CAG
K  M  S  V  S  K  A  E  P  V  M  L  E  Q  I  S  R  I  S  Q
2345/321
CAG ATC GGC TAT TAT CTG CAT CGC GCC AGT ATG CGC GGT AGC GGC GTG TTG TTA AGC CGC
Q  I  G  Y  Y  L  H  R  A  S  M  R  G  S  G  V  L  L  S  R
2405/341
GAA CTG CAT CCC GTC GCG CCG TTG TTA GAT AAC CTG ATT TCT GCG CTA AAT AAA GTT TAT
E  L  H  P  V  A  P  L  L  D  N  L  I  S  A  L  N  K  V  Y
2465/361
CAG CGT AAA GGG GTG AAT ATC AGT ATG GAT ATT TCA CCA GAA ATC AGT TTT GTC GGC GAG
Q  R  K  G  V  N  I  S  M  D  I  S  P  E  I  S  F  V  G  E
2525/381
CAA AAC GAC TTT GTC GAA GTG ATG GGC AAC GTA CTG GAC AAC GCT TGT AAA TAT TGT CTG
Q  N  D  F  V  E  V  M  G  N  V  L  D  N  A  C  K  Y  C  L
2585/401
GAG TTT GTC GAG ATT TCG GCT CGC CAG ACC GAC GAT CAT TTG CAT ATT TTC GTC GAA GAT
E  F  V  E  I  S  A  R  Q  T  D  D  H  L  H  I  F  V  E  D
2645/421
GAC GGC CCA GGC ATT CCC CAC AGC AAA CGT TCC CTG GTG TTT GAT CGC GGT CAG CGC GCC
D  G  P  G  I  P  H  S  K  R  S  L  V  F  D  R  G  Q  R  A
2705/441
GAT ACC CTA CGA CCA GGA CAA GGC GTG GGG CTG GCT GTC GCG CGC GAG ATT ACG GAA CAA
D  T  L  R  P  G  Q  G  V  G  L  A  V  A  R  E  I  T  E  Q
2765/461
TAC GCC GGG CAG ATC ATT GCC AGC GAC AGT CTG CTC GGT GGC GCC CGT ATG GAG GTC GTT
Y  A  G  Q  I  I  A  S  D  S  L  L  G  G  A  R  M  E  V  V
2825/481
TTT GGC CGA CAG CAT CCC ACA CAG AAA GAG GAA TAA
F  G  R  Q  H  P  T  Q  K  E  E  *
← phoQ ends

```

Figure 29. Construction of the suicide vector for introducing $\Delta P_{phoPQ-107::TT}$ *araC* P_{BAD} *phoPQ* deletion-insertion mutation.

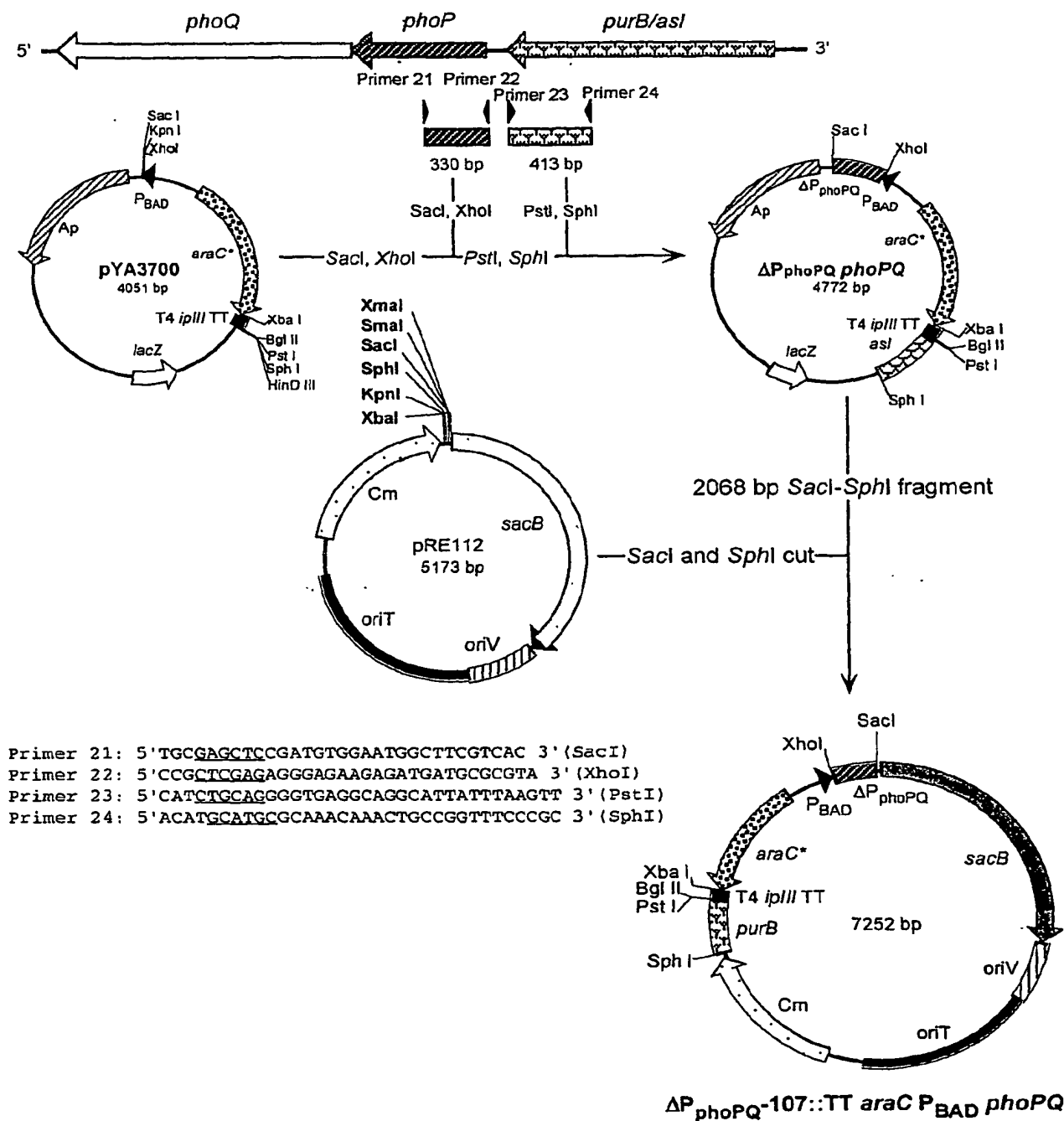
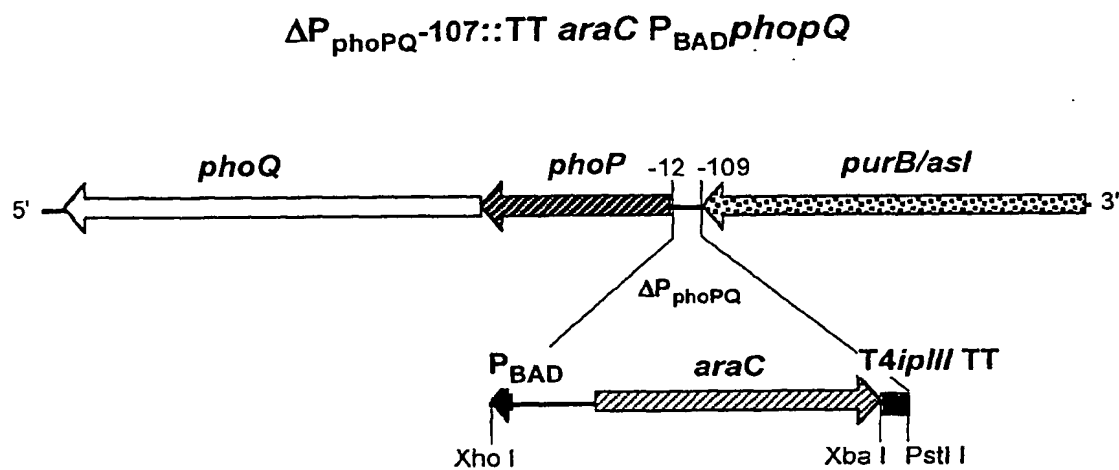
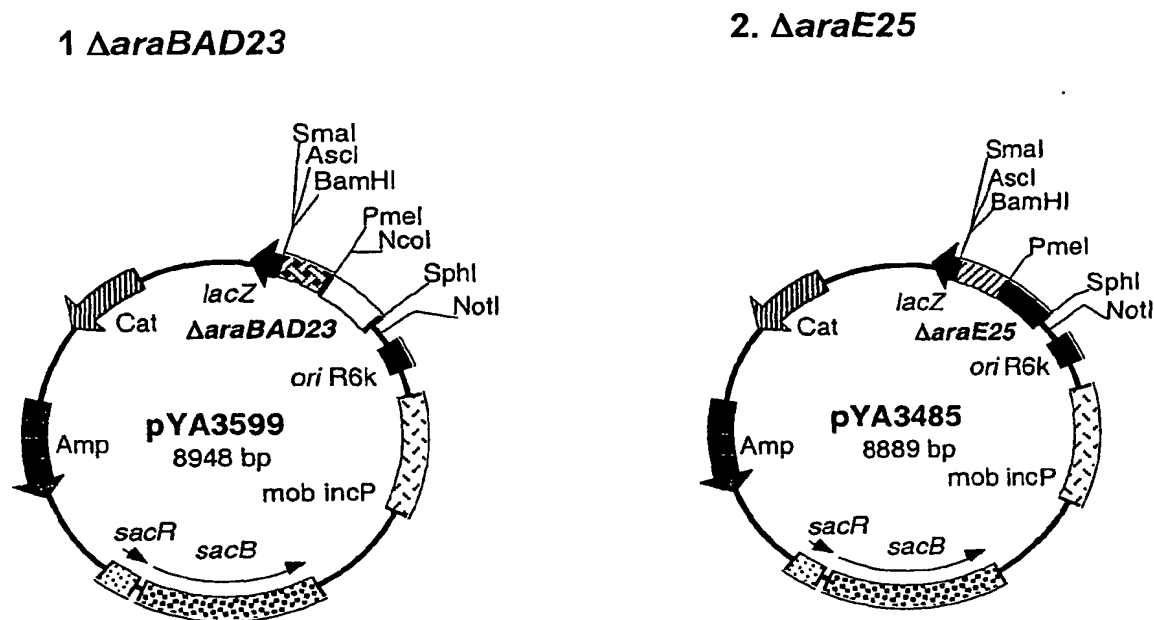


Figure 30. Chromosomal map of $\Delta P_{phoPQ-107::TT}$ *araC* P_{BAD} *phoPQ* deletion-insertion mutation.



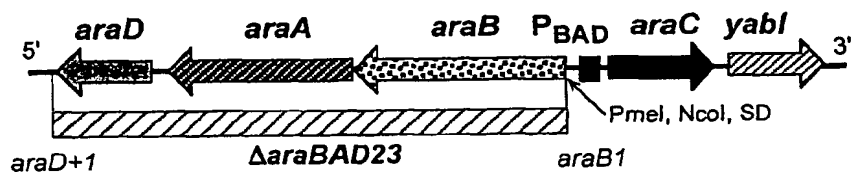
phoPQ promoter region (-12 to -109) deleted and 1344 bp P_{BAD} *araC* TT inserted.

Figure 31. Diagrams of the suicide vectors for introducing the $\Delta araBAD23$ and $\Delta araE25$ deletion mutations.



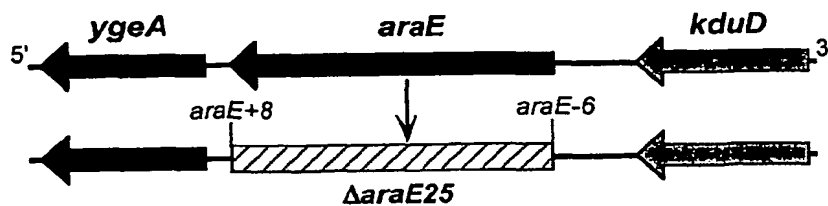
In *Salmonella* chromosome:

1. $\Delta araBAD23$



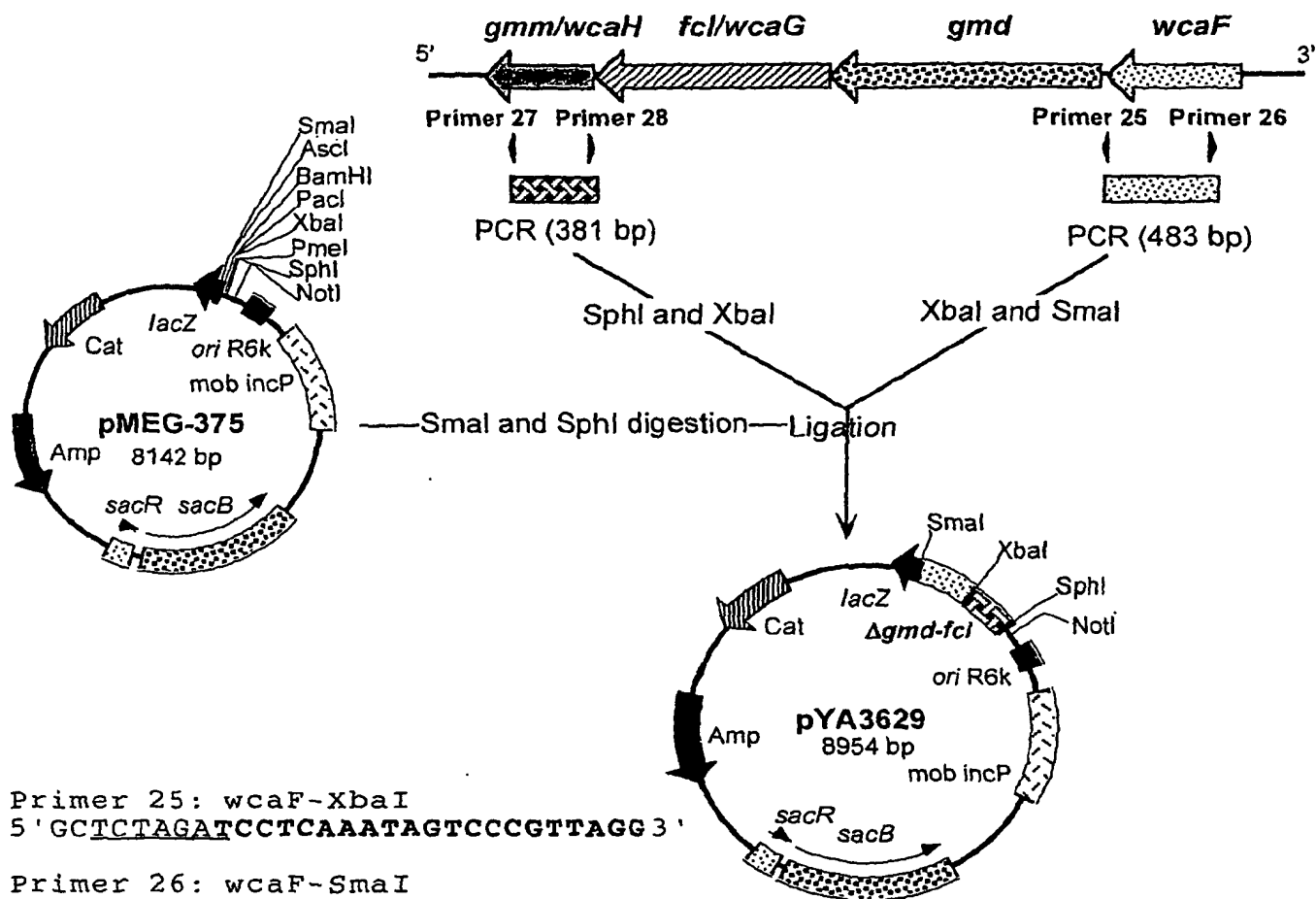
$araBAD$ ($araB1$ to $araD+1$, total of 4101 bp) deleted and SD, NcoI, and PmeI sites generated

2. $\Delta araE25$



$araE-6$ to $araE+8$, total of 1433 bp was deleted

Figure 32. Construction of the suicide vector for introducing the $\Delta(gmd-fcl)$ -26 deletion mutation.



gmm/wcaH: Guanosine di-P mannose mannosyl hydrolase
fcl/wcaG: Colanic acid gene cluster, bifunctional GDP fucose synthetase
gmd: Fucose biosynthesis; GDP-D-mannose 4,6-dehydratase
wcaF: Involved in lipopolysaccharide biosynthesis, putative acyltransferase

Figure 33. Chromosomal map of the $\Delta(gmd-fcl)$ -26 deletion mutation.

In *Salmonella* chromosome:

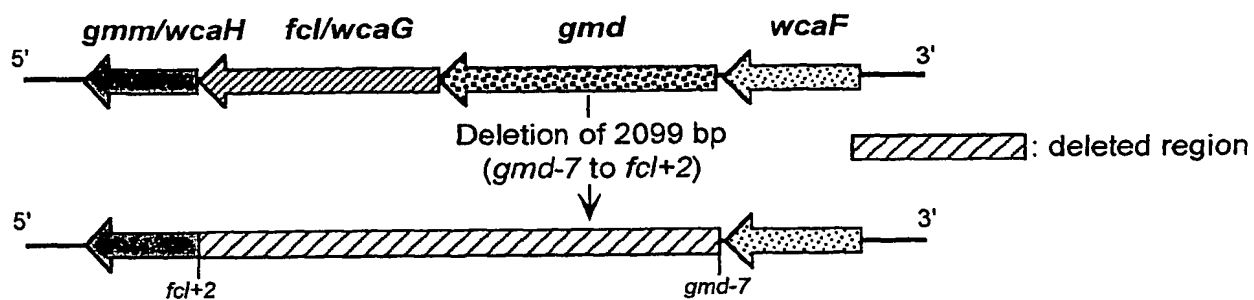


Figure 34. Diagrams of all the suicide vectors listed in Table 2.

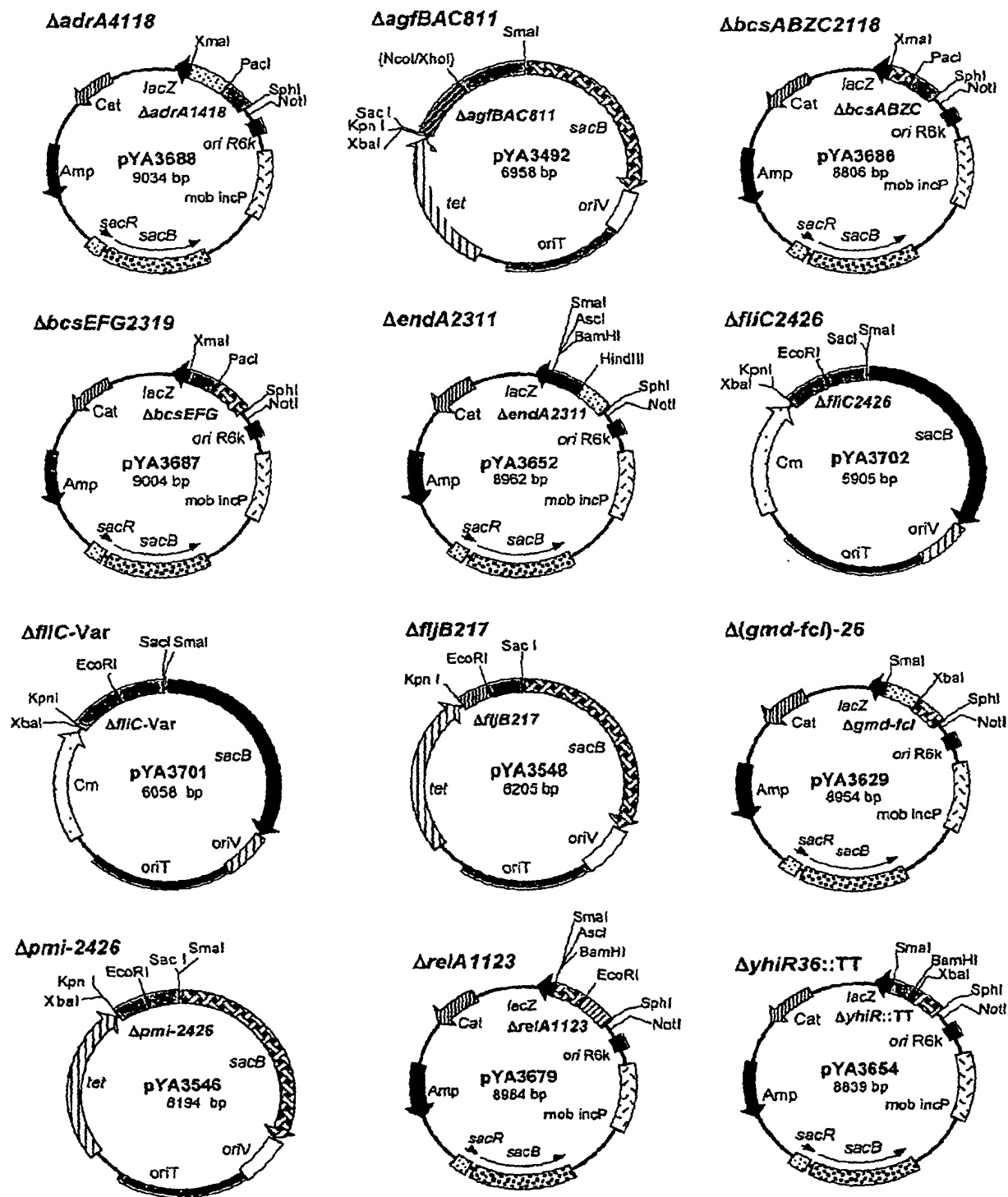
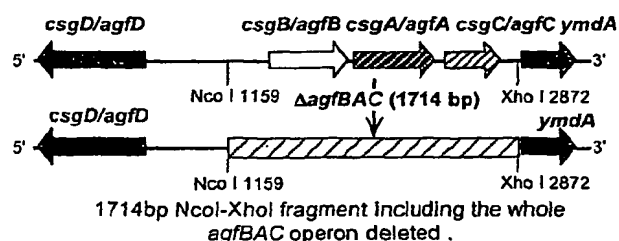
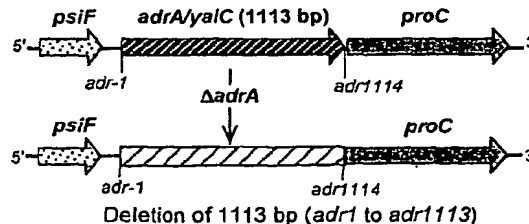
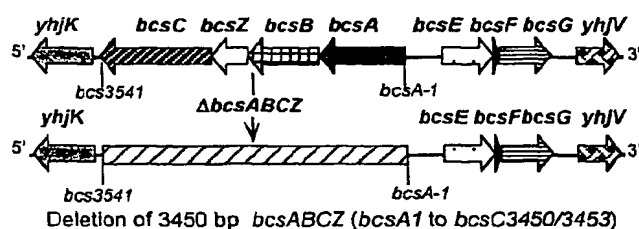
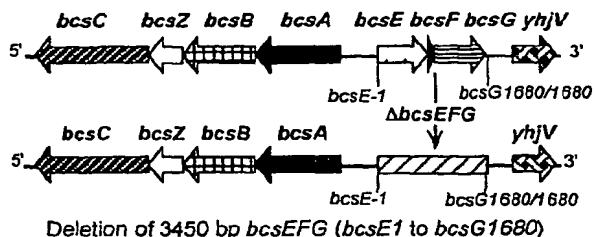
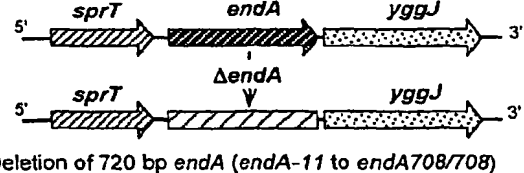
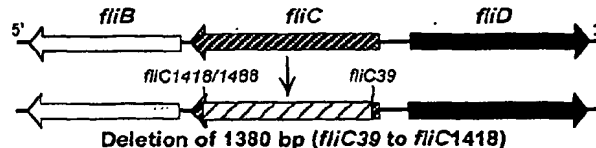
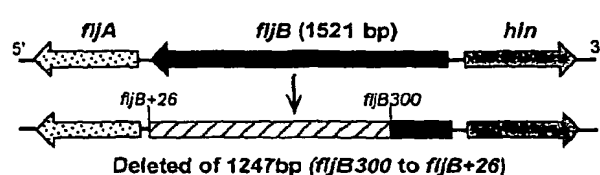
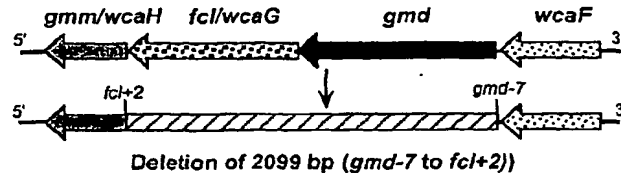
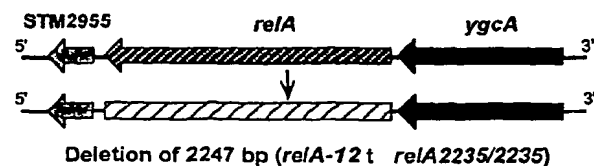
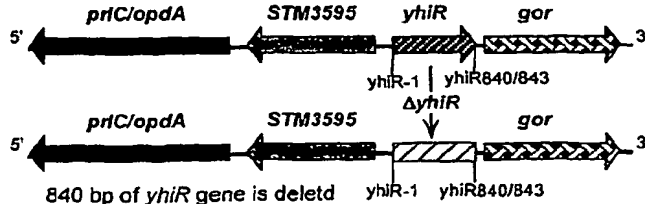


Figure 35. Deletion mutations after insertion into *Salmonella* chromosome. **Δ agfBAC811** **Δ adrA1418** **Δ bcsABZC2118** **Δ bcsEFG2319** **Δ endA2311** **Δ fliC825** **Δ fliB217** **Δ (gmd-fcl)-26** **Δ relA1123** **Δ yhiR36::TT**

: deleted region

Figure 36. DNA and amino acid sequences of *sopB* and the flanking region of *S. typhimurium* chromosome.

GGA ATA GGA AAA ACG AAT ATT CTT CGT CAC GGT CTT ACT TGT CCG GGG CTT TGC TGG CAT
 S Y S F R I N K T V T K S T R P K A P M
 ←STM1092 starts ↑

ACA CAC ACC TGT ATA ACA TTT GAT GTA ACG CCG TTA CTT TAC GCA GGA GTA AAT CGG TGA
 SD (STM1092)
 ATT TGA TCT GAG TCA AGA AGG TGG GTT TTC AAT AAA AGT TGT GCC ATA AAT TGT GAA GTT
 TGT AGA TTT TAT GAA CAT TTG ATG TAC CGA TCT CCC CCA TGA TCG CCA CTA CGT ATG GAC
 GTC AGG ATG CCT CCC CGC CTG ATC AGA AGC GTT TCC TCA TTA AAA AGG ACA TTT TTT TAA
 AGT TCC TGG TGC ATA AAA GTC ACA TCC TTT TAA AGG GTT GTT AAC CCT GTT GAA TGT TCC
 SD
 CAC TCC CCT ATT CAG GAA TAT TAA AAA CGC T
 ↑SD-*sopB* deleted (*sopB*-18 to *sopB*1686)

sopB starts →

1/1	31/11
ATG CAA ATA CAG AGC TTC TAT CAC TCA GCT	TCA CTA AAA ACC CAG GAG GCT TTT AAA AGC
M Q I Q S F Y H S A	S L K T Q E A F K S
61/21	91/31
CTA CAA AAA ACC TTA TAC AAC GGA ATG CAG	ATT CTC TCA GGC CAG GGC AAA GCG CCG GCT
L Q K T L Y N G M Q	I L S G Q G K A P A
121/41	151/51
AAA GCG CCC GAC GCT CGC CCG GAA ATT ATT	GTC CTG CGA GAA CCC GGC GCG ACA TGG GGG
K A P D A R P E I I	V L R E P G A T W G
181/61	211/71
AAT TAT CTA CAG CAT CAG AAG GCG TCT AAC	CAC TCG CTG CAT AAC CTC TAT AAC TTA CAG
N Y L Q H Q K A S N	H S L H N L Y N L Q
241/81	271/91
CGC GAT CTT CTT ACC GTC GCG GCA ACC GTT	CTG GGT AAA CAA GAC CCG GTT CTA ACG TCA
R D L L T V A A T V	L G K Q D P V L T S
301/101	331/111
ATG GCA AAC CAA ATG GAG TTA GCC AAA GTT	AAA GCG GAC CGG CCA GCA ACA AAA CAA GAA
M A N Q M E L A K V	K A D R P A T K Q E
361/121	391/131
GAA GCC GCG GCA AAA GCA TTG AAG AAA AAT	CTT ATC GAA CTT ATT GCA GCA CGC ACT CAG
E A A A K A L K K N	L I E L I A A R T Q
421/141	451/151
CAG CAG GAT GGC TTA CCT GCA AAA GAA GCT	CAT CGC TTT GCG GCA GTA GCG TTT AGA GAT
Q Q D G L P A K E A	H R F A A V A F R D
481/161	511/171
GCT CAG GTC AAG CAG CTT AAT AAC CAG CCC	TGG CAA ACC ATA AAA AAT ACA CTC ACG CAT
A Q V K Q L N N Q P	W Q T I K N T L T H
541/181	571/191
AAC GGG CAT CAC TAT ACC AAC ACG CAG CTC	CCT GCA GCA GAG ATG AAA ATC GGC GCA AAA
N G H H Y T N T Q L	P A A E M K I G A K
601/201	631/211
GAT ATC TTT CCC AGT GCT TAT GAG GGA AAG	GGC GTA TGC AGT TGG GAT ACC AAG AAT ATT
D I F P S A Y E G K	G V C S W D T K N I

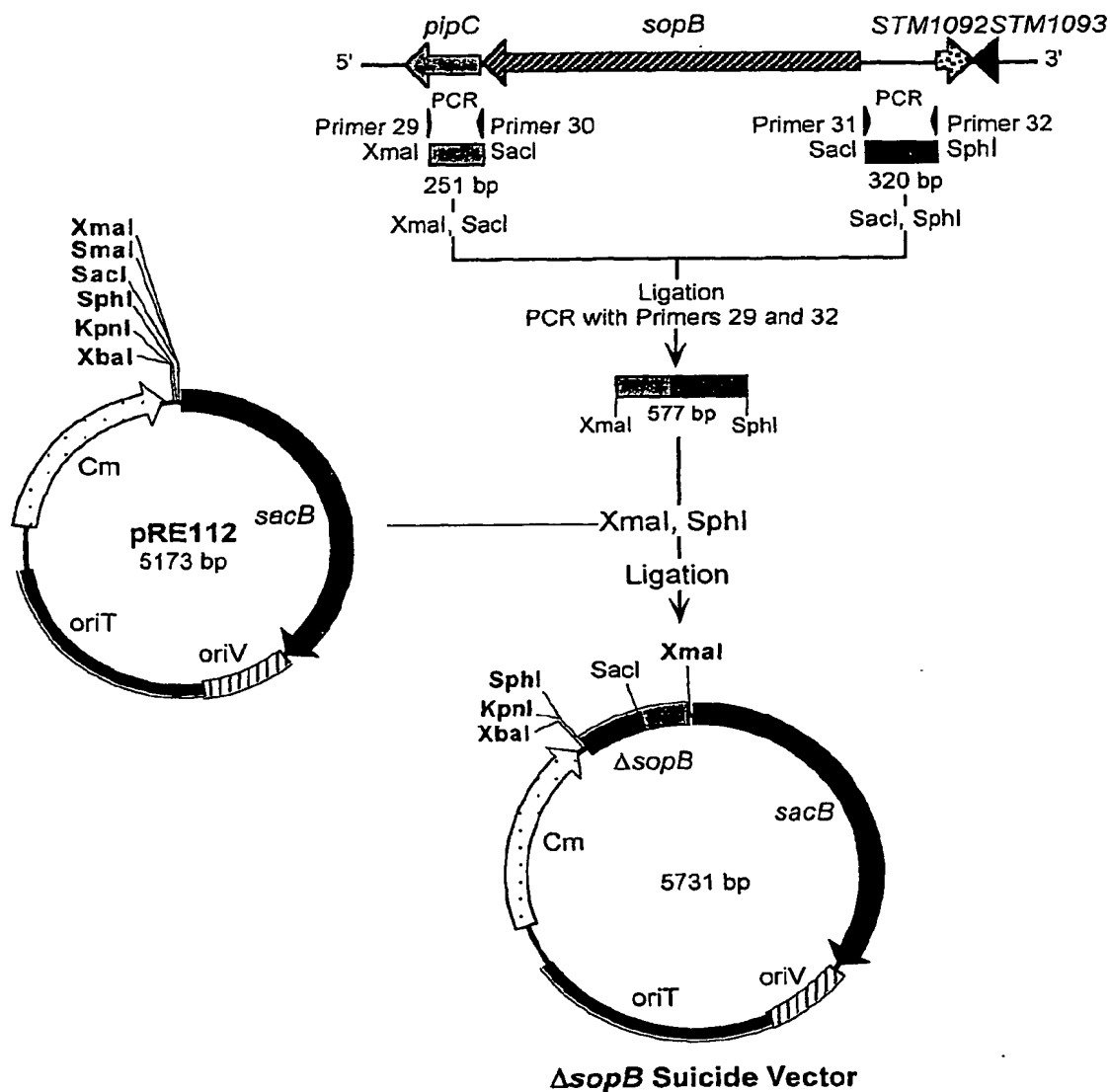
Figure 36. (cont'd,

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661/221                               691/231
CAT CAC GCC AAT AAT TTG TGG ATG TCC ACG GTG AGT GTG CAT GAG GAC GGT AAA GAT AAA
H  H  A  N  N  L  W  M  S  T  V  S  V  H  E  D  G  K  D  K
721/241                               751/251
ACG CTT TTT TGC GGG ATA CGT CAT GGC GTG CTT TCC CCC TAT CAT GAA AAA GAT CCG CTT
T  L  F  C  G  I  R  H  G  V  L  S  P  Y  H  E  K  D  P  L
781/261                               811/271
CTG CGT CAC GTC GGC GCT GAA AAC AAA GCC AAA GAA GTA TTA ACT GCG GCA CTT TTT AGT
L  R  H  V  G  A  E  N  K  A  K  E  V  L  T  A  A  L  F  S
841/281                               871/291
AAA CCT GAG TTG CTT AAC AAA GCC TTA GCG GGC GAG GCG GTA AGC CTG AAA CTG GTA TCC
K  P  E  L  L  N  K  A  L  A  G  E  A  V  S  L  K  L  V  S
901/301                               931/311
GTC GGG TTA CTC ACC GCG TCG AAT ATT TTC GGC AAA GAG GGA ACG ATG GTC GAG GAC CAA
V  G  L  L  T  A  S  N  I  F  G  K  E  G  T  M  V  E  D  Q
961/321                               991/331
ATG CGC GCA TGG CAA TCG TTG ACC CAG CCG GGA AAA ATG ATT CAT TTA AAA ATC CGC AAT
M  R  A  W  Q  S  L  T  Q  P  G  K  M  I  H  L  K  I  R  N
1021/341                             1051/351
AAA GAT GGC GAT CTA CAG ACG GTA AAA ATA AAA CCG GAC GTC GCC GCA TTT AAT GTG GGT
K  D  G  D  L  Q  T  V  K  I  K  P  D  V  A  A  F  N  V  G
1081/361                             1111/371
GTT AAT GAG CTG GCG CTC AAG CTC GGC TTT GGC CTT AAG GCA TCG GAT AGC TAT AAT GCC
V  N  E  L  A  L  K  L  G  F  G  L  K  A  S  D  S  Y  N  A
1141/381                             1171/391
GAG GCG CTA CAT CAG TTA TTA GGC AAT GAT TTA CGC CCT GAA GCC AGA CCA GGT GGC TGG
E  A  L  H  Q  L  L  G  N  D  L  R  P  E  A  R  P  G  G  W
1201/401                             1231/411
GTT GGC GAA TGG CTG GCG CAA TAC CCG GAT AAT TAT GAG GTC GTC AAT ACA TTA GCG CGC
V  G  E  W  L  A  Q  Y  P  D  N  Y  E  V  V  N  T  L  A  R
1261/421                             1291/431
CAG ATT AAG GAT ATA TGG AAA AAT AAC CAA CAT CAT AAA GAT GGC GGC GAA CCC TAT AAA
Q  I  K  D  I  W  K  N  N  Q  H  H  K  D  G  G  E  P  Y  K
1321/441                             1351/451
CTC GCA CAA CGC CTT GCC ATG TTA GCC CAT GAA ATT GAC GCG GTA CCC GCC TGG AAT TGT
L  A  Q  R  L  A  M  L  A  H  E  I  D  A  V  P  A  W  N  C
1381/461                             1411/471
AAA AGC GGC AAA GAT CGT ACA GGG ATG ATG GAT TCA GAA ATC AAG CGA GAG ATC ATT TCC
K  S  G  K  D  R  T  G  M  M  D  S  E  I  K  R  E  I  I  S
1441/481                             1471/491
TTA CAT CAG ACC CAT ATG TTA AGT GCG CCT GGT AGT CTT CCG GAT AGC GGT GGA CAG AAA
L  H  Q  T  H  M  L  S  A  P  G  S  L  P  D  S  G  G  Q  K
1501/501                             1531/511
ATT TTC CAA AAA GTA TTA CTG AAT AGC GGT AAC CTG GAG ATT CAG AAA CAA AAT ACG GGC
I  F  Q  K  V  L  L  N  S  G  N  L  E  I  Q  K  Q  N  T  G
1561/521                             1591/531
GGG GCG GGA AAC AAA GTA ATG AAA AAT TTA TCG CCA GAG GTG CTC AAT CTT TCC TAT CAA
G  A  G  N  K  V  M  K  N  L  S  P  E  V  L  N  L  S  Y  Q
1621/541                             1651/551
AAA CGA GTT GGG GAT GAA AAT ATT TGG CAG TCA GTA AAA GGC ATT TCT TCA TTA ATC ACA
K  R  V  G  D  E  N  I  W  Q  S  V  K  G  I  S  S  L  I  T
1681/561
TCT TGA GTCTTGAGGTAACATAT  ATG GAA AGT CTA TTA AAT CGT TTA TAT GAC GCG TTA GGC
S  * ↑          SD          M  E  S  L  L  N  R  L  Y  D  A  L  G
(sopB1686)                pipC starts

```

Figure 37. Construction of the suicide vector for introducing the $\Delta sopB$ deletion mutation into the *Salmonella* chromosome.



Primer 29: 5' TTCCCCCGGGGCAGTATTGTCTGCGTCAGCG 3' (*XmaI*-N)
 Primer 30: 5' TTGAGAGCTCGTCTTGAGGTAACTATATGGAAAG 3' (*SacI*-N)
 Primer 31: 5' TTGAGAGCTCGAATAGGGGAGTGGGAACATTC 3' (*SacI*-C)
 Primer 32: 5' ACATGCATGCGGCATACACACACCTGTATAACA 3' (*SphI*-C)

Figure 38. Chromosomal map of Δ *sopB* deletion mutation.

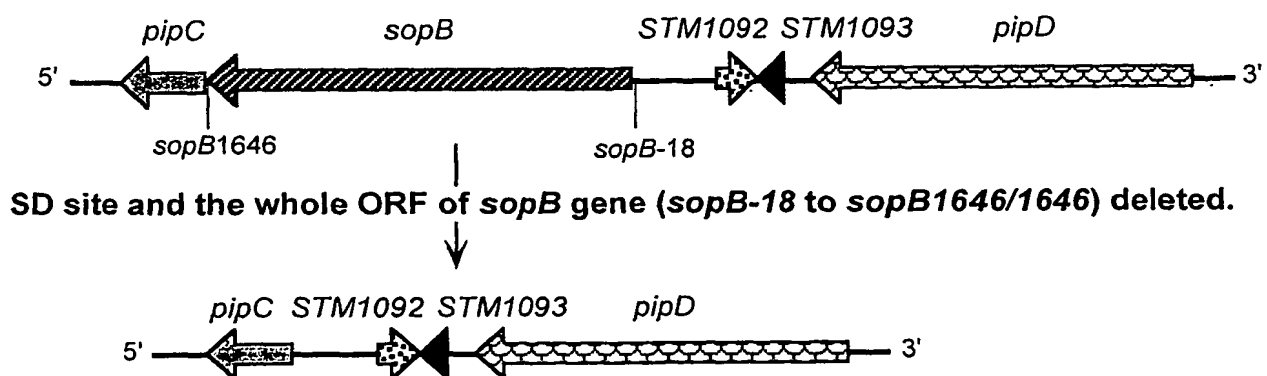
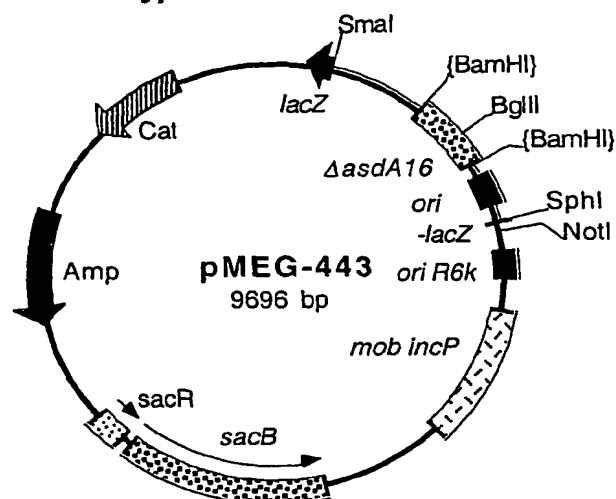


Figure 39. Diagrams of the suicide vectors for introducing $\Delta asdA16$ into *S. typhimurium* and $\Delta asdA25$ into *S. paratyphi* A and *S. typhi* strains.

$\Delta asdA16$ for *S. typhimurium*



$\Delta asdA25$ for *S. paratyphi* A and *S. typhi*

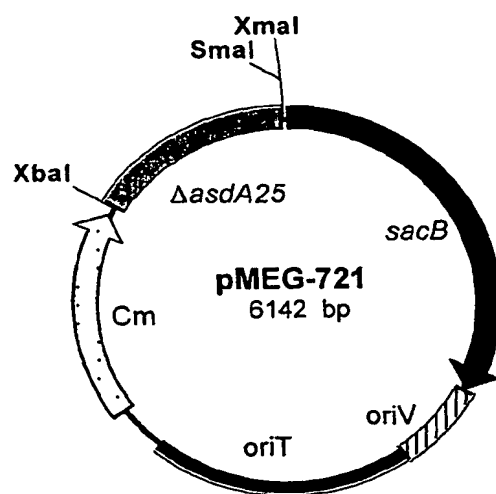
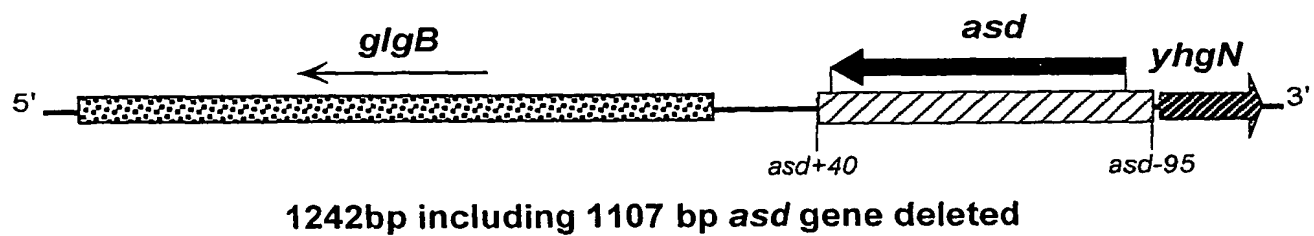
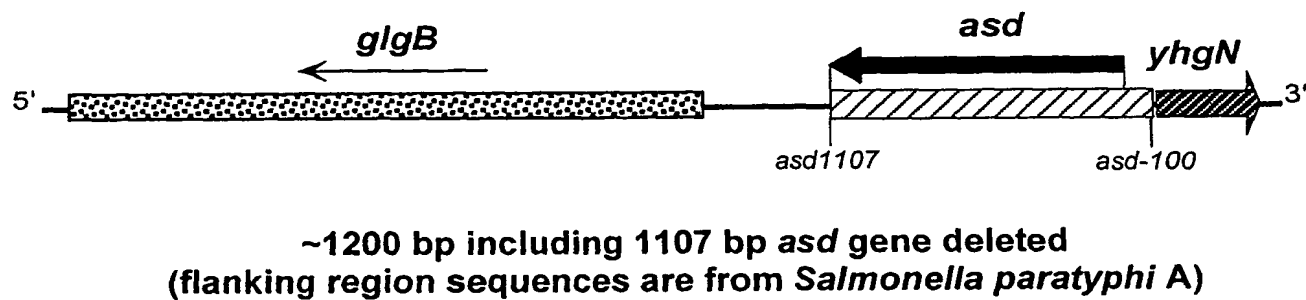


Figure 40. Chromosomal maps of $\Delta asdA16$ and $\Delta asdA25$ deletion mutation.

$\Delta asdA16$ for *S. typhimurium*



$\Delta asdA25$ for *S. paratyphi A* and *S. typhi*



: deleted region

Figure 41. *Asd*⁺ vectors with pSC101, p15A, pBR and pUC origins of replication to regulate plasmid copy numbers.

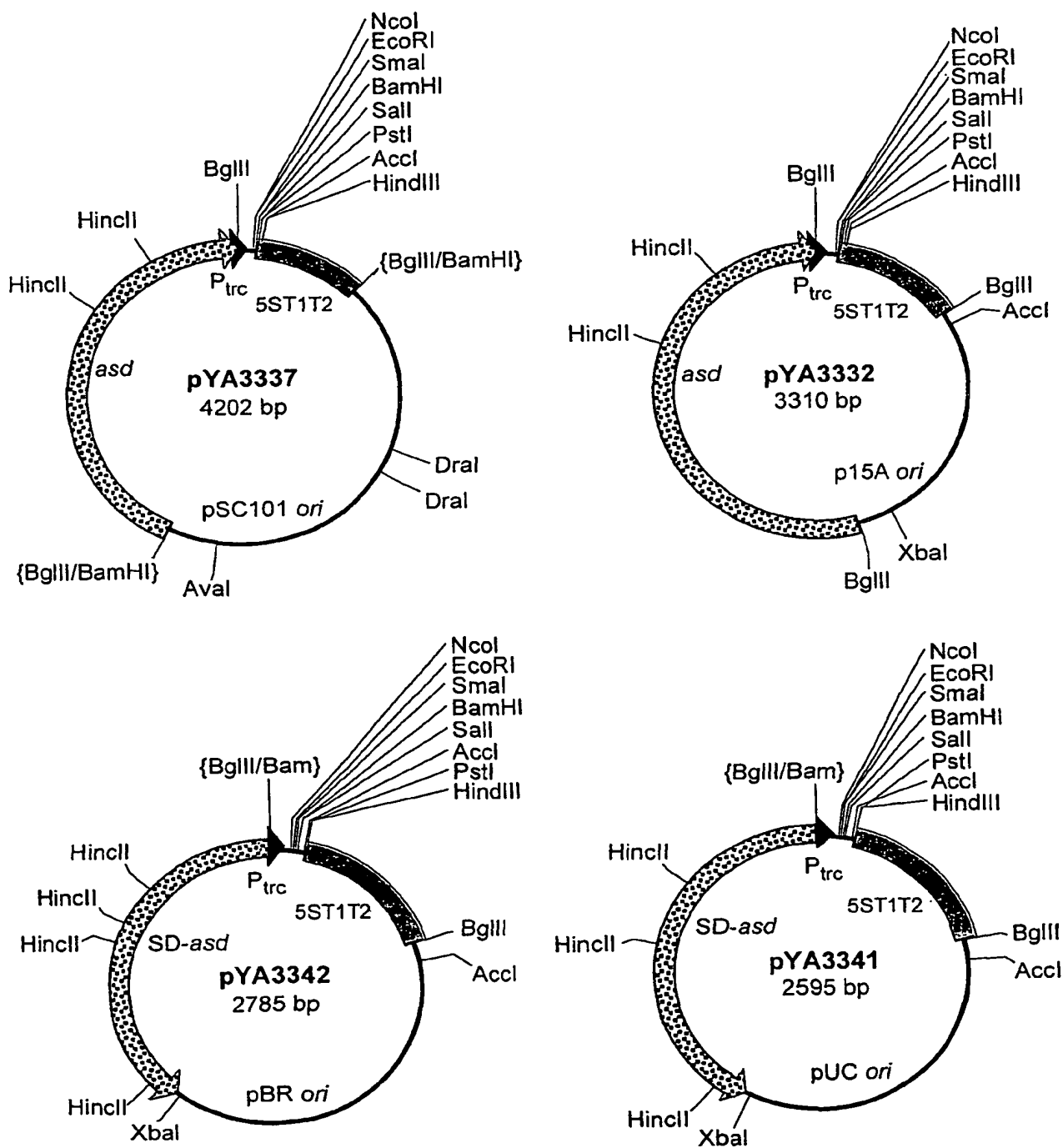


Figure 42. Nucleotide sequence of P_{trc} and the multiple cloning sites (MCS) of Asd⁺ vectors in Figure 41.

```

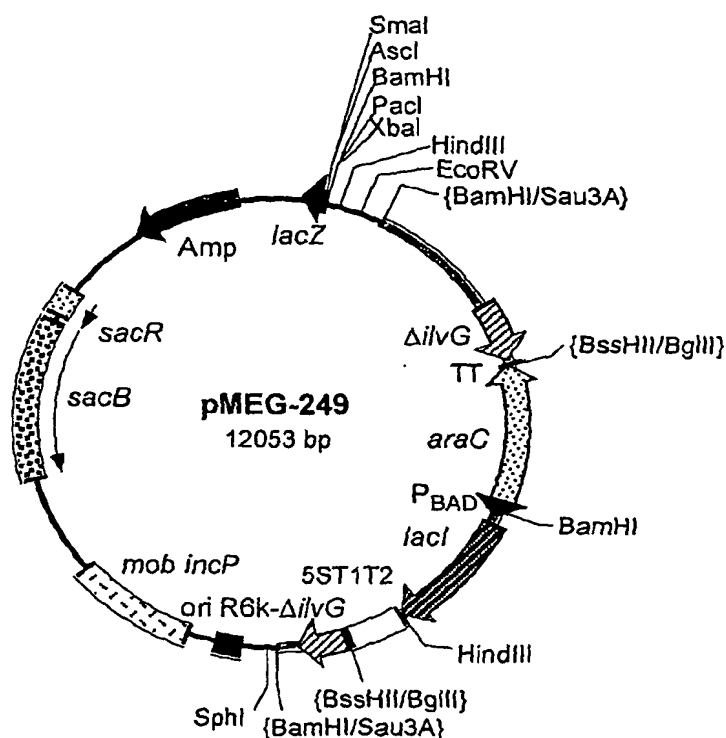
                -35                                -10
ATT CTG AAA TGA GCT GTT GAC AAT TAA TCA TCC GGC TCG TAT AAT GTG TGG AAT TGT

GAG CGG ATA ACA ATT TCA CAC AGG AAA CAG ACC ATG GAA ATT CGC AAT TCC CGG GGA
                SD                                NcoI                                SmaI
                Met Gly Ile Arg Asn Ser Arg Gly

BamHI                PstI
TCC GTC GAC CTG CAG CCA AGC TCC CAA GCT T
                Sali                                HindIII
Ser Val Asp Leu Gln Pro Ser Ser Gln Ala

```

Figure 43. Diagram of the suicide vector for introducing $\Delta ilvG3::TT araC P_{BAD} lacI$ TT deletion-insertion mutation and map of $\Delta ilvG3::TT araC P_{BAD} lacI$ TT mutation in the *Salmonella* chromosome.



In *Salmonella* chromosome:

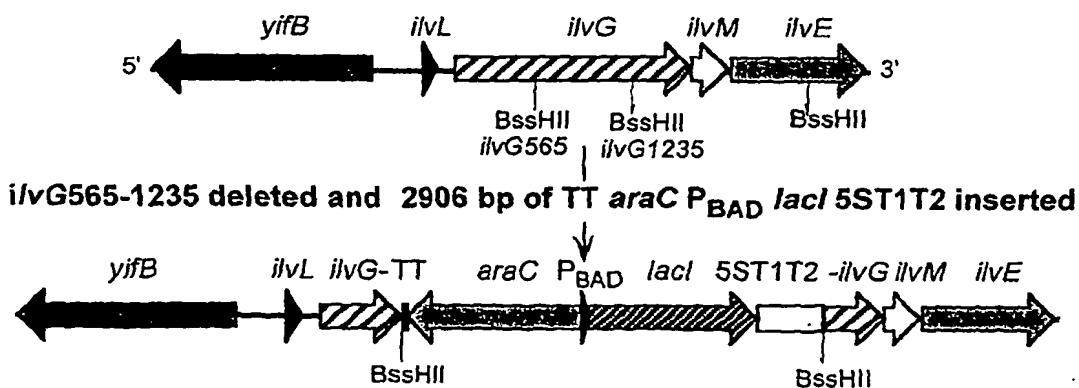


Figure 44: Nucleotide and amino acid sequences of *S. typhimurium fimH* and FimH protein

```

1/1                               31/11
atg aaa ata tac tca gcg cta ttg ctg gcg ggg acc gcg ctc ttt ttc acc cat ccc gcg
M K I Y S A L L L A G T A L F F T H P A
61/21                               91/31
ctg gcg acg gtt tgc cgt aat tca aac ggg acg gcg acc gat atc ttt tac gac ctg tca
L A ↑ T V C R N S N G T A T D I F Y D L S
121/41                               151/51
gat gtt ttc acc agc ggc aat aat cag ccg gga cag gtg gtg acg ctg ccg gaa aaa tca
D V F T S G N N Q P G Q V V T L P E K S
181/61                               211/71
ggg tgg gtc ggc gta aac gcg acg tgc ccg gcg ggg aca acg gtg aat tat acc tac cga
G W V G V N A T C P A G T T V N Y T Y R
241/81                               271/91
agc tat gta tca gaa tta ccg gta caa agt acc gaa gga aat ttt aaa tac ctc aag ttg
S Y V S E L P V Q S T E G N F K Y L K L
301/101                               331/111
aat gac tac ctt ctg ggc gcg atg agc atc acc gat agt gtc gct ggc gta ttt tat ccg
N D Y L L G A M S I T D S V A G V F Y P
361/121                               391/131
ccc cgt aac tat att ctc atg ggc gtc gac tat aac gtg tcg cag caa aag ccg ttt ggc
P R N Y I L M G V D Y N V S Q Q K P F G
421/141                               451/151
gtg cag gac tca aag ctg gtt ttt aaa tta aaa gtg ata cgg cct ttt att aat atg gtg
V Q D S K L V F K L K V I R P F I N M V
481/161                               511/171
acg atc cct cgc cag aca atg ttt acc gtc tat gtg acg acc tct acc ggc gac gcg ttg
T I P R Q T M F T V Y V T T S T G D A L
541/181                               571/191
agc acg ccg gta tat acc att agc tac agc ggc aaa gtg gaa gtg ccg caa aac tgt gaa
S T P V Y T I S Y S G K V E V P Q N C E
601/201                               631/211
gtg aat gcc gga cag gtc gtg gag ttt gat ttc gcc gat atc gcc gcg tcg tta ttt agt
V N A G Q V V E F D F G D I G A S L F S
661/221                               691/231
cag gcg ggg gcg ggt aat cgt ccg caa ggc gtc acg ccg caa acg aaa acc att gct atc
Q A G A G N R P Q G V T P Q T K T I A I
721/241                               751/251
aaa tgt acc aac gtc gcg gcg cag gcc tat tta tcg atg cgg ctt gaa gcc gaa aag gcc
K C T N V A A Q A Y L S M R L E A E K A
781/261                               811/271
tca ggg cag gcg atg gtg tcc gat aat ccg gat tta ggc ttt gtg gtt gct aat agc aac
S G Q A M V S D N P D L G F V V A N S N
841/281                               871/291
ggg acg ccg ctt aca ccc aat aat ttg tcg agt aaa att ccg ttt cat ctt gat gat aac
G T P L T P N N L S S K I P F H L D D N
901/301                               931/311
gcc gcc gct cgc gta ggt att cgc gcc tgg cca atc agc gtg acg ggg att aaa ccg gcg
A A A R V G I R A W P I S V T G I K P A
961/321                               991/331
gaa ggg ccg ttt act gcg cgc ggc tat cta cga gtc gat tat gat taa
E G P F T A R G Y L R V D Y D *

```

Amino acids 1-22 constitute the signal sequence cleaved from the mature protein (amino acids 23 to 335). Arrow indicates site of signal peptidase cleavage.

Figure 45: Construction of *fimH* *Asd*⁺ vectors

Primer1: *FimH*-*Nco*I (starting with amino acid 23)

5'- CAT GCC ATG GCA TGC ACG GTT TGC CGT AAT TCA AAC G-3'

Primer2: *FimH*₁₀₀-*Hind*III (starting with amino acid 122)

5'-GCC CAA GCT TA TTA ACG GGG CGG ATA AAA TAC GCC AGC-3'

Primer3: *FimH*-*Hind*III (starting with terminal codon)

5'- GCC CAA GCT TTT AAT CAT AAT CGA CTC GTA GAT AGC C-3'

